THE UNIVERSITY OF ALBERTA

PELLETED RATIONS FOR SHEEP: EFFECTS ON FEED CONSUMPTION AND UTILIZATION, VOLATILE FATTY ACID PRODUCTION AND FERMENTATION RATE IN THE RUMEN

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ABSTRACT

Experiments were conducted to study the effects of pelleting of lowland hay rations on rate of gain and efficiency of feed utilization of lambs, rate of rumen fermentation, relative production and proportions of volatile fatty acids in rumen fluid, and cellulose digestion in the rumen.

The results of feeding trials with group-fed and individually-fed lambs, showed that pelleted lowland hay rations resulted in marked increases in feed consumption and rate of gain, and improved efficiency of feed utilization, as compared to similar non-pelleted rations. Lowland hay was improved in feeding value as a result of pelleting and gave excellent results in fattening lambs at levels as high as 84 per cent of pelleted rations. Digestibility of dry matter, crude protein and gross energy was lower in rations containing lowland hay than in one composed of alfalfa and barley; digestibility decreased with increasing levels of lowland hay in the ration. The effect of pelleting on digestibility depended on the level of lowland hay in the ration; digestibility was decreased at low levels of lowland hay, but increased at high levels of lowland hay in the ration.

An <u>in vitro</u> study of fermentation rates in rumen samples indicated that rate of fermentation decreased with increasing levels of lowland hay in the ration, but was not affected by pelleting of the rations. Differences in rate of fermentation, as measured by production of gas and acid in manometric fermentations, did not reflect differences noted in feed intake or digestibility when lambs were fed similar pelleted and non-pelleted rations.

Concentrations of volatile fatty acids in rumen fluid decreased as the level of lowland hay in the ration was increased. The proportion

of acetic acid increased and the proportions of propionic and/or butyric acids decreased with higher levels of lowland hay in the ration. Pelleting of the rations increased the concentrations of volatile fatty acids with rations containing low levels of lowland hay, but had no effect with rations containing high levels of lowland hay; the proportions of volatile fatty acids in rumen fluid were not affected by pelleting of the rations. Concentrations and proportions of volatile fatty acids in rumen samples did not reflect differences in feed intake or rate of gain of lambs.

Cellulose digestion in the rumen was measured by an *in vivo* technique. A higher percentage of cellulose was digested when lambs were fed non-pelleted lowland hay rations than when lambs were fed an alfalfa-barley ration. Pelleting of a ration containing 60 per cent of lowland hay decreased cellulose digestion, whereas pelleting of a ration containing 90 per cent of lowland hay increased cellulose digestion.

The effect of increased energy and available nitrogen on utilization of lowland hay was tested in feeding trials. The addition of stabilized animal fat increased the energy content of pelleted and non-pelleted rations, but did not improve rate of gain or efficiency of feed utilization. Supplementation of rations containing high levels of lowland hay with urea and amino acids did not improve feed intake or rate of gain of lambs.

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INTRODUCTION

In recent years much emphasis has been placed on methods for improving efficiency of livestock production. As a result, notable advances have been made, particularly in the areas of swine and poultry production. While progress with ruminants has been less spectacular, advances in other areas have stimulated research on various aspects relating to efficiency of production of ruminants. A method which has created widespread interest, and shows considerable promise as a means of improving efficiency, is the pelleting of rations for ruminants. Marked increases in gain, improved efficiency of feed use, and economies in labour requirements have been attributed to the use of pelleted rations.

The process followed in pelleting of rations containing roughage consists of grinding and mixing the ingredients, and forcing the mixture through a perforated metal die by application of pressure. Prior to pelleting the material may be conditioned with steam. The feed is compressed into pellets which may be cubes, rectangles or cylinders, varying in size from 1/16 to one inch in diameter and from about 3/16 to two inches in length. Size and shape of pellets depends upon the die selected and the adjustment of the knife.

The nutritive value of a ration depends on level of feed intake as well as on nutrient composition, and it is apparent that an important result of the pelleting process is to modify the feed in such a way that voluntary feed consumption is increased. Fine grinding of the feed increases the rate of passage through the digestive tract, permitting increased feed intake; greater palatability of the ground feed, as a result of pelleting, increases voluntary consumption. It has also been suggested that pelleting of the ration may result in a faster rate of fermentation in the rumen, greater production of volatile fatty acids and more efficient utilization of energy from the volatile fatty acids produced. These in

turn may lead to an increase in the nutritive value of the ration.

The greatest improvement in nutritive value as a result of pelleting occurs when lower quality roughages are used. Low quality roughage, that would normally be refused or consumed in small amounts by sheep and cattle may be readily consumed with very little wastage when incorporated into a pelleted ration. When properly supplemented to overcome deficiencies of minerals, protein and energy, low quality feeds may be used to excellent advantage.

Native lowland hay forms a large part of the roughage fed to ruminants in many areas of Western Canada. This hay, comprised principally of slough grass (Beckmannia syzigachne), sedges (Carex sp.) and blue grasses (Poa sp.), is usually coarse, fibrous and low in protein content, because of the advanced stage of growth obtained before soil conditions permit harvesting.

Since experimental evidence was lacking on the effects of pelleting of low quality forages on rate of digestion, rate of fermentation and production of volatile fatty acids, experiments were undertaken to determine the effects of pelleting of lowland hay rations for lambs on rate of gain, efficiency of feed utilization, rate of rumen fermentation and relative production of volatile fatty acids in the rumen.

REVIEW OF LITERATURE

A. Historical Background

Although "pressed" feed was used for cavalry horses by the French army as early as 1860 (Tillman, 1961), the use of pelleted rations for livestock has only recently become popular. The process first came into use commercially about thirty years ago in preparation of plant

protein supplements. It was next used to process dehydrated grass and alfalfa, which were marketed in cubed or pelleted form, either alone or in combination with cereals. Eventually the process was extended to complete rations for poultry, swine and ruminants.

The use of pelleted rations for poultry gained quick acceptance. In addition to advantages such as ease of storing and handling, reduced dustiness and decreased feed wastage, as compared with mash, it was noted in several studies that pelleted rations resulted in increased rate of growth and improved efficiency of feed utilization (Patton et al., 1937; Heywang and Morgan, 1944; Ziegenhagen et al., 1947; Bearse et al., 1952).

Advantages, similar to those for poultry, have been reported with pelleted rations for swine (Steffen, 1953; Lehrer and Keith, 1953; Dinusson and Bolin, 1958), but the adoption of pelleted feeds for swine has not become widespread. Increased deposition of fat in the carcass of swine fed pelleted rations has prevented extensive use of the process (Bowland, 1956).

Interest in the use of pelleted rations for ruminants has developed mainly during the past fifteen years. An earlier report by Powell (1938), which indicated that feeding of checkered or cubed feeds to dairy cows depressed the fat-test in milk, discouraged the use of pelleted rations for dairy cattle. The first indication that pelleting of rations for ruminants might be beneficial was a report by Ensminger et al. (1948), who noted an increase in rate of gain of steers fed pelleted rations. A later report by Neale (1953), that pelleted low-quality alfalfa hay was equal or superior to high-quality, long alfalfa hay in feeding value for lambs, stimulated a marked interest in pelleted rations for ruminants. As a result, many studies have been conducted on the use of pelleted rations for sheep and cattle.

Some information is now available on many aspects of the use of pelleted rations for ruminants. The effects of pelleting on rate of gain, feed conversion and carcass quality of fattening lambs and cattle have been studied. The influence of pelleting on composition of the ration, palatability and rate of passage of feed through the digestive tract, digestibility of nutrients, rumen production of volatile fatty acids, and the papillae of the rumen epithelium has also received attention. Experimental results pertaining to each aspect will be presented in this review.

B. Effects of Pelleting on Rate of Gain, Feed Conversion and Carcass Quality

The observation that pelleting of rations resulted in increased gain of steers and lambs has led to many studies on the effects of pelleting on rate of gain, efficiency of feed conversion and carcass quality of ruminants. Studies with lambs have indicated that pelleting of the ration results in increased feed intake, higher rate of gain, improved feed conversion, and similar or superior carcass quality as compared to lambs fed non-pelleted rations (Esplin et al., 1957; Weir et al., 1957; Hogue, 1958; Bowstead, 1959; Loosli, 1959). Similar but less spectacular results have been obtained in experiments with beef cattle (Weir et al., 1959; Kercher and Hilston, 1958; Wallace and Hubbert, 1959). The greatest benefits from pelleting have been obtained with rations containing low-quality roughages (Neale, 1953; Cate et al., 1955; Jordan, 1959; Reynolds and Lindahl, 1960).

While many studies have shown advantages for pelleting, others have indicated disadvantages. Reduced feed consumption and decreased rate of gain of steers and heifers fed pelleted rations have been reported

(Baker et al., 1955; Baker et al., 1957; Cullison, 1961).

It soon was recognized that the level of roughage in the ration exerted an influence on the response obtained when feed was pelleted. In studies with lambs it was shown that when the proportion of roughage in the ration was low, pelleting had little or no beneficial effect, and in some instances depressed feed intake and rate of gain (Hartman et al., 1959; Jordan, 1959; McClure et al., 1960). When the proportion of roughage in the ration was high, pelleted rations tended to increase feed consumption and rate of gain as compared to non-pelleted rations (Hartman et al., 1959; Jordan, 1959; Thomas et al., 1960). Similar effects have been noted in studies with beef cattle. Beardsley et al. (1959) reported that the response to pelleting increased as the level of roughage in the ration was increased from 30 to 60 per cent.

The optimum response from pelleting appears to occur with rations containing approximately 70 per cent of roughage. Neale (1955) and Neale (1958) reported that the response of lambs to pelleted rations containing 70 per cent of roughage was greater than with rations containing 50, 60 or 80 per cent of roughage. Hopson et al. (1960) concluded that 50 and 30 per cent were maximum and minimum levels respectively of concentrate that would produce maximum utilization of rations by lambs. Kercher and Hilston (1958) obtained as great an increase in feed intake and rate of gain of steers with a ration containing 50 per cent of alfalfa as with one containing 70 per cent. Weir et al. (1959) found no difference in rate of gain of lambs or steers fed pelleted rations containing 70 or 100 per cent of high-quality alfalfa.

In some studies, the increase in rate of gain noted with animals fed pelleted rations has been attributed to the increase in feed intake which occurs. Esplin et al. (1957) and Meyer et al. (1959a) observed that

lambs fed pelleted rations consumed more feed and gained at a faster rate than those fed non-pelleted rations; however, when equal amounts of similar pelleted and non-pelleted rations were fed, no differences in rate of gain were noted. Meyer et al. (1959b) found that pelleted rations showed no advantage when data on rate of gain were adjusted for feed intake. The results suggest that a close association exists between level of feed intake and rate of gain.

C. Effects of Pelleting on Composition of the Ration

Esplin et al. (1957) found no differences in dry matter, ether extract, ash, crude fibre, nitrogen or nitrogen-free extract, when rations were pelleted. In some reports, pelleting resulted in a small decrease in crude fibre (Dinusson and Bolin, 1958), accompanied by a slight increase in ether extract (Lindahl and Davis, 1955; Lindahl and Reynolds, 1959). The increase in ether extract noted did not result in an increase in gross energy, which suggested that it probably represented non-fat ether-soluble materials released as a result of cellular rupture. It seems evident that pelleting of rations has little effect on the content of major nutrients.

The pelleting process may have some effect on the starch components of a ration. Jensen et al. (1959) found that starch in pelleted corn was more susceptible to malt amylase digestion than starch in ground corn. Hastings and Miller (1961) reported that pressure used in the pelleting process increased total solubles and soluble starch in grain, and did not destroy inherent enzymes. Salsbury et al. (1961) noted that boiling in excess water, or heating in an autoclave, resulted in hydration and more rapid digestion of starch. Since the pelleting process involves steam treatment and the application of heat, the changes which were noted may occur to some extent in pelleted feeds.

D. Effects of Pelleting on Palatability and Rate of Passage of Feed Through the Digestive Tract

It is recognized that improved performance of ruminants when fed pelleted rations is associated with a higher level of feed intake; however, the reason why voluntary feed consumption is increased is not clear. It seems to be related, in part at least, to the palatability of the ration. Meyer *et al*. (1959b) observed that feed intake and rate of gain of lambs were decreased when alfalfa was finely ground. Moistening the ground feed with water increased feed consumption and daily gains, although not as much as when the feed was pelleted. Since the addition of water increased feed consumption, they concluded that fine grinding was a major factor in permitting increased feed intake, and that pelleting merely improved palatability of a dusty feed. Lloyd *et al*. (1960a) obtained similar results. Wallace *et al*. (1961), in studies on utilization of native meadow hay, concluded that pelleting increased palatability of the forage.

Although palatability or acceptability of a feed may be an important factor, it appears that voluntary feed consumption is regulated principally by rate of digestion and rate of passage of feed through the digestive tract (Crampton, 1957; Lambourne, 1957). Vandersall (1960) reported that ration digestibility strongly influenced feed intake. Consumption was related to quality of the ration; any process which increased rate of digestion increased feed consumption. Blaxter *et al*. (1961) concluded that "sheep eat to a constant distension of their digestive tracts measured by the 'fill', which is in turn determined by the rate of passage of the food and its digestibility". Blaxter *et al*. (1956) reported maximum voluntary consumption of 1800 grams per day by wethers fed long hay, compared to 2400 grams per day when hay was ground and cubed.

Increased intake was associated with increased rate of passage, allowing more feed to be consumed to fill the digestive tract.

Other studies have shown that the consistency of rumen contents may influence rate of passage of food. Balch (1950) observed that when long hay was fed to cows, the rumen contents consisted of a tightly packed mass of dry, fibrous material above a very fluid layer. Subsequent regurgitation and rumination reduced particle size, enabling mixing of the ingesta with the fluid contents of the rumen, and passage through the reticulo-omasal orifice to the omasum. When ground hay was fed, the rumen contents had a thick, even consistency. Newly arrived feed was retained longer in the reticulum and anterior rumen, in position to pass the reticulo-omasal orifice. A high initial rate of excretion from the rumen occurred with ground hay, whereas reduction in particle size of long hay was necessary before passage occurred.

In other studies it was reported that cubed or pelleted rations also resulted in rumen contents of a thick, even consistency (Blaxter and Graham, 1956; Meyer et al., 1959a). Balch (1958b) found that cubes or pellets had almost completely disintegrated by the time they reached the reticulo-rumen, resulting in consistency of the rumen contents similar to that with ground hay. Balch (1950) and Balch et al. (1955) found that, although ground hay caused a faster initial rate of excretion of ingesta from the rumen, the total time of passage was more prolonged than when long hay was fed. Total weight and total dry matter of the rumen contents were decreased, allowing thorough mixing of newly arrived feed with that remaining from previous meals, and prolonging total time of excretion.

Fineness of grinding and level of intake of feed affect rate of passage through the digestive tract. Blaxter et al. (1956) compared rations of long, medium ground (1/4 inch screen) and pelleted, and finely ground

(twice through 1/16 inch screen) and pelleted hay, each fed to sheep at three levels of intake. Rate of passage of food from the reticulo-rumen increased with intake and with fineness of grinding of the feed. Similar results were reported by Blaxter and Graham (1956). When sheep were fed long hay, the gut contained feed residues corresponding to 2.6 days' food intake immediately before the next meal, compared to about half this amount when finely ground and cubed hay was fed (Blaxter et al., 1956). Meyer et al. (1959a) concluded that pelleted roughages resulted in faster digestion, possibly because more surface area of the finer particles allows greater microbial attack of cellulose. The accelerated digestion promoted faster passage of feed from the reticulo-rumen, and resulted in increased feed intake.

E. Effects of Grinding and Pelleting on Digestibility of the Ration

The observation that palatability of the ration, and rate of passage of feed through the digestive tract may be affected by treatment of the ration, led to studies on the effects of grinding and pelleting on digestibility of the various components of the ration. In some studies it was indicated that the physical form of the ration had marked effects on digestibility. Blaxter and Graham (1956) reported that grinding and pelleting of roughage resulted in depressed digestibility of all components except ether extract. The cellulosic fraction was mainly affected; crude fibre digestibility decreased 40 per cent, while digestibility of nitrogen-free extract decreased only 12 per cent. Fecal losses of energy were increased; pelleted rations were 27 per cent lower than chopped material in apparent digestibility of gross energy. Net energy was the same for both chopped and pelleted hays. Increased fecal losses were balanced by reduced methane production, because of decreased fermentation

of cellulose, and lower heat losses, which were attributed to less muscular effort required in prehension, chewing and rumination when sheep were fed pelleted hay. Swanson and Herman (1952) noted that feeding of ground roughage decreased apparent digestibility of crude fibre, and Lloyd et al. (1960b) observed that digestible energy decreased when chopped hay was ground, and that pelleting of the ground hay caused a further decrease. Reynolds and Lindahl (1960) reported that ground and pelleted rations depressed digestibility of dry matter and crude fibre. They attributed the depression to inability of the animals to select the more nutritious portions of the feed when it was ground or pelleted. Long et al. (1955) found lower digestion coefficients of dry matter, crude fibre, crude protein and nitrogen-free extract when the ration was ground, but noted that pelleting of the ground ration restored digestion coefficients to the level of the original ration.

Level of intake and fineness of grinding of the ration also affect digestibility. Blaxter et al. (1956) observed that digestibility of the ration decreased with increasing level of intake and increasing fineness of grind, reflecting decreased time spent in the digestive tract. Rate of decrease in digestibility with level of intake was greater when food passed quickly through the tract (finely ground and cubed), than when it remained in the digestive tract for longer periods (long hay). Rodrigue and Allen (1960) reported similar results from feeding of ground rations.

In some studies it has been reported that grinding and pelleting of rations has little effect on digestibility. Meyer et al. (1959a) found no difference between chopped or pelleted alfalfa hay in apparent digestibility of dry matter, total digestible nutrients, gross energy, digestible energy, metabolizable energy, net energy or methane production. They reported a faster rate of breakdown of holocellulose in the rumen when

pelleted rations were fed to lambs. Other reports (Esplin et al., 1957; Weir et al., 1959; Hopkins et al., 1960) have indicated no differences in digestion coefficients of dry matter, crude protein or nitrogen-free extract, but small reductions in digestibility of crude fibre, as a result of pelleting.

The level of concentrates in rations appears to have marked effects on digestibility. Several reports have indicated that increasing levels of concentrates in pelleted and non-pelleted rations result in increased digestibility of dry matter, crude protein, nitrogen-free extract, and energy, but decreased digestibility of crude fibre and cellulose (Balch et al., 1955; Balch, 1957; Brent et al., 1961). It was suggested that higher levels of concentrates in the ration increased the amount of starch and allowed more rapid growth of starch-digesting micro-organisms at the expense of cellulolytic organisms, resulting in increased digestion of crude protein and nitrogen-free extract, but decreased digestion of fibre.

F. Effects of the Ration on Rumen Volatile Fatty Acids

The presence of volatile fatty acids[(1)] in rumen contents was known as early as 1883 (Annison and Lewis, 1959), but information on individual fatty acids present, and their concentrations, was not available until Elsden (1946) devised a method for separation of acetic, propionic and butyric acids in mixtures of VFA. Later, using improved methods of separation, Gray et al. (1952) demonstrated the presence of formic, acetic, propionic, n-butyric, isobutyric, valeric, caproic and heptoic acids in rumen fluid of sheep. Acetic acid made up 62 to 70 per cent and propionic acid 16 to 27 per cent of the VFA. Annison (1954) reported the presence

[(1)]Referred to hereafter as VFA in the text of this thesis.

of isovaleric and 2-methylbutyric acids, and concluded that formic acid and acids of longer chain length than valeric (referred to hereafter as "higher acids") were present in trace amounts only, if at all, under normal feeding conditions.

It was soon recognized that VFA make a major contribution to the energy requirements of the ruminant. McAnally and Phillipson (1942) and Barcroft et al. (1944) showed that considerable quantities of VFA were absorbed from the rumen. Carroll and Hungate (1954) calculated that, depending on the ration fed, from 5000 to 12,000 Calories of energy became available daily from VFA produced in the rumen of cattle. Phillipson and Cuthbertson (1956) calculated that 600 - 1200 Calories of energy were absorbed daily as VFA from the rumen of sheep. The total energy turnover of fasting adult cattle and sheep was estimated at 6500 and 1100 Calories per day respectively.

The observation that VFA are important sources of energy for the ruminant led to many studies which indicated that the relative proportions of VFA produced in the rumen appear to have considerable influence on ruminant metabolism. Armstrong and Blaxter (1957a) reported that fasting animals made efficient use of mixtures of VFA. Acetic acid fed alone resulted in a heat increment of 41 per cent of the calories metabolized, compared to 13 per cent for propionic acid, 16 per cent for butyric acid and 17 per cent for a mixture of the three acids in the proportions of 5:3:2. Acetic acid when fed alone, was not rapidly metabolized, causing depressed rumen pH, accumulation of blood VFA and increased nitrogen excretion. Armstrong et al. (1957) found that very small quantities of propionic and butyric acids facilitated utilization of acetic acid. A mixture of acetic, propionic and butyric acids in the proportions of 90:6:4 resulted in a heat increment of 15 per cent. They concluded that

acetic acid fed alone increased nitrogen excretion because of breakdown of body tissue to provide energy, but that mixtures of the acids were efficiently used for energy and saved body protein from oxidation.

Very different results were obtained when individual VFA were administered to fattening animals (Armstrong and Blaxter, 1957b). Heat increments were higher, particularly for propionic and butyric acids. Heat increments of 67, 44 and 38 Calories per 100 Calories metabolized were obtained for acetic, propionic and butyric acids respectively. Efficiency of conversion of Calories for fat deposition was low, with considerable amounts of VFA being oxidized to provide energy for fat synthesis. Armstrong et al. (1958) reported that efficiency of utilization of mixtures of VFA, administered to fattening animals, was the sum of the effects of the acids given singly. A heat increment of 68 per cent was obtained with a mixture of acetic, propionic, and butyric acids in the proportions of 75:15:10. Increasing propionic and butyric acids to provide a mixture of the same three acids in the proportions of 25:45:30, resulted in a heat increment of 41 to 42 per cent.

The physical nature of the ration and proportions of concentrates have marked effects on relative production of VFA in the rumen. It has been reported that when rations containing high levels of long hay were fed, acetic was the predominant acid produced, with smaller amounts of propionic and butyric acids (Gray et al., 1952; McCarthy et al., 1958). In other studies it was noted that rations low in hay and high in concentrates resulted in decreased proportions of acetic acid and increased proportions of propionic acid (Balch et al., 1955; Balch and Rowland, 1957; Leffel, 1960). Balch and Rowland (1957) reported that decreasing roughage from 100 per cent to 1/3 of the ration, reduced acetic acid levels in the rumen from 69 per cent to 62 per cent and increased propionic acid

from 18 to 20 per cent of the total VFA. When rations containing only eight per cent of roughage were fed, acetic acid in rumen liquid decreased from 69 to 40 - 45 per cent and propionic acid increased from 18 to 31 - 39 per cent of the total VFA.

Other studies have demonstrated that grinding the roughage may affect the proportions of various VFA. Balch and Rowland (1957) noted that rations containing 50 per cent of ground roughage depressed acetic acid production to about 50 per cent, and increased propionic acid to about 30 per cent of the total VFA. Balch (1958a) noted that the percentage of acetic acid decreased about 13 per cent and propionic acid increased about 10 per cent in rumen liquid of cows fed rations containing 50 per cent of ground roughage, as compared to similar rations with long hay.

There is little evidence available on the effects of pelleting on the production of VFA. Ensor et al. (1959) found a diet of ground and pelleted hay plus heated concentrates had marked effects on relative proportions of VFA in the rumen, and was highly effective in increasing gain and efficiency of gain of fattening steer calves. Rations containing only ground and pelleted hay had small effects on relative proportions of VFA in the rumen, whereas the addition of heated corn produced a marked decrease in the proportion of acetic acid, and increased the proportions of propionic, butyric and valeric acids. Increasing the amount of heated corn, at the expense of ground and pelleted roughage, resulted in much greater changes in the proportions of VFA. Shaw et al. (1960) obtained similar results on the proportions of acetic and propionic acids in rumen liquid of steers fed rations containing ground and pelleted hay and flaked corn. Rate of gain and efficiency of gain increased 22 and 15 per cent respectively, compared to rations of chopped hay and ground corn.

Composition of the ration also appears to influence the concentrations of VFA in the rumen. Balch and Rowland (1957) reported only minor fluctuations between meals in concentrations of VFA in rumen liquid of cows fed all-roughage rations. This suggested a fairly steady rate of fermentation. Feeding rations with only 44 per cent of roughage resulted in marked fluctuations in VFA concentrations; a high concentration occurred three to six hours after feeding, and a low concentration just before feeding. Greater fluctuations occurred when cows were fed rations containing 8 per cent of long roughage or 50 per cent of ground roughage. They concluded that rations containing ground roughage, or high proportions of concentrates, promoted rapid fermentation in the rumen three to six hours after feeding.

While the production of a number of VFA has been shown to occur in the rumen, and their importance in energy metabolism has been indicated, it is difficult to estimate the rate of production of each, or the total amount of VFA produced. The complexity of rumen fermentations, dilution of rumen contents, absorption of VFA and passage of rumen material to the omasum constitute sources of variability that cast doubt on the validity of values derived for total VFA produced. A number of studies (McCarthy et al., 1957; McCarthy et al., 1958; Brown et al., 1960; Leffel, 1960) have indicated that absorption of VFA is directly related to the rate of production. Consequently, analysis of rumen contents appears to give, within limits, a useful measure of relative rates of production of the various acids.

G. Effects of Pelleted Rations on the Rumen Epithelium

Pelleting of the ration, because of its effects on production of VFA, may have some influence on papillary development in the rumen

epithelium. Flatt *et al*. (1959) and Warner *et al*. (1959) reported that papillary growth was stimulated by chemical end-products of rumen fermentation, particularly butyric and propionic acids. Kunkel *et al*. (1959) found a significant positive correlation between voluntary feed consumption and length, width and density of rumen papillae. However, pelleted feeds appeared to have only slight effects on average papillary length.

In some instances, pelleted rations have been associated with deleterious effects on the rumen epithelium, increasing the occurrence of a condition known as parakeratosis or hyperkeratinization. This was first described (Jensen *et al*., 1958) as a non-contagious disease of sheep, characterized by hardening, enlargement and clumping of rumen papillae. Excessive layers of keratinized nucleated squamous epithelial cells accumulated on the papillae. Rate of gain was reduced and as many as 43 per cent of the lambs fed pelleted rations were affected. Hopkins *et al*. (1960) reported parakeratosis of the rumen in lambs fed non-pelleted rations, but the incidence was low. A similar condition, with long, dark and unhealthy appearing papillae and areas of keratinous tissue, has been reported in steers fed pelleted and non-pelleted rations (Beardsley *et al*., 1959; Cullison, 1961; Garrett *et al*., 1961).

Beardsley *et al*. (1959) reported that dark-colored rumen walls occurred in ruminants fed high-roughage pellets, but the condition did not appear to be related to parakeratosis. The real significance of the discoloration was not fully understood.

EXPERIMENTS AT THE UNIVERSITY OF ALBERTA

Experiments were conducted from 1959 to 1961 to study the effects in sheep of pelleting rations containing varying levels of native lowland hay on:

1. Rate of gain and efficiency of feed utilization.
2. Rate of rumen fermentation.
3. Relative production and proportions of volatile fatty acids in the rumen.
4. Rate of cellulose digestion in the rumen.

I. Effects of Pelleting and Level of Lowland Hay on Rate of Gain and Efficiency of Feed Utilization

Status of the Problem

Large quantities of native lowland hay are used for livestock production in Western Canada; however, the low quality of much of the hay limits its value as a livestock feed. Lowland hays are usually harvested at an advanced stage of growth because of wet soil conditions. As a consequence, the forage is usually coarse, fibrous and low in protein content. When fed to sheep or cattle, consumption is usually much lower than when upland grass or legume hays are fed. Reduced feed intake, combined with low quality of the forage, often leads to unsatisfactory results when native lowland hay is fed.

The nutritive value of a ration depends on level of feed intake as well as on nutrient composition. Therefore, any method that would improve palatability of lowland hay might enhance its value as a feed for livestock. Since pelleting of rations containing high levels of roughage has been shown to increase feed intake, rate of gain and feed

efficiency, it seemed desirable to determine the effects of pelleting on the nutritive value of native lowland hays for lambs.

Experimental (General)

The same lot numbers were used in each of the four trials conducted and denote the physical form in which the ration was fed. The control ration (Lot 1) consisted of long alfalfa and whole barley hand-fed in the proportions of 1:2. Lots 2, 4 and 6 were fed non-pelleted rations containing varying proportions of lowland hay, barley and supplements. The hay was chopped into one inch to two inch lengths; barley was coarsely ground in Trial 1 and rolled in succeeding trials. Lots 3, 5 and 7 were fed pelleted rations of the same composition as those fed to Lots 2, 4 and 6 respectively. In preparing the pelleted rations, the roughage and concentrate were ground through a 1/8 inch screen in a hammermill, mixed, and processed into 9/64 inch pellets(1). Ten per cent of alfalfa was included in all rations containing lowland hay because of its beneficial effect on ruminant digestion.

The native lowland hays were comprised mainly of slough grass (*Beckmannia syzigachne*), sedges (Carex sp.) and blue grasses (Poa sp.). Wild barley (*Hordeum jubatum*) was also an important species in the lowland hay fed in the second trial.

Analyses of feed and fecal samples for dry matter and crude protein were conducted by A.O.A.C. (1960) methods. Gross energy of feed and fecal samples was determined with a Parr Oxygen Bomb Calorimeter. Samples of the rations were obtained and analyzed at 14 day intervals in each trial; averages of chemical analyses of dry matter, crude protein and gross energy for each ration are shown in Tables 2, 4, 6 and 8. Variations in analyses of crude protein between similar non-pelleted and pelleted rations may be attributed to sampling errors. Results of chemical analyses(2) for crude

(1)Sprout Waldron Ace Pellet Mill.
(2)All data on feed analyses in this thesis are reported on oven-dry bases.

protein and gross energy in the roughages and barley used in each trial are shown in Table 1.

Table 1. Crude Protein and Gross Energy in Roughages and Barley Used in Experimental Rations

	Native Lowland Hay		Alfalfa		Barley	
Trial	Crude Protein (%)	Gross Energy (Therms/lb.)	Crude Protein (%)	Gross Energy (Therms/lb.)	Crude Protein (%)	Gross Energy (Therms/lb.)
1	11.2	2.11	16.5	1.99	11.5	1.94
2	11.4	1.94	18.3	1.93	13.3	1.95
3	8.9	2.04	15.9	1.97	13.3	1.97
4	11.6	2.00	18.1	1.99	11.0	1.98

In the first three trials, lambs were divided into comparable groups on the basis of numbers, breed and weight. Water was freely available to the sheep in all trials. Lambs were weighed at the beginning of Trials 1 to 3, and at 14 day intervals thereafter until marketed. Weights were obtained at weekly intervals in Trial 4. Chilled carcass weights and grades were obtained on all lambs marketed.

Analyses of variance of the data were conducted as outlined by Goulden (1956) and significant mean differences were determined by Duncan's Multiple Range Test as outlined by Duncan (1955) and extended by Kramer (1956) for unequal subclass numbers.

Trial 1

Object

To determine the effects of pelleting rations containing varying levels of native lowland hay and supplemental animal fat on rate of gain and efficiency of feed utilization of fattening lambs.

Experimental

Fifty-six wether lambs of range type (mainly Rambouillet and Romnelet breeding) were divided into seven comparable lots of eight lambs each. The formulation of the rations fed the different lots is shown in Table 2.

Table 2. Rations for Trial 1

Lot Number	1	2	3	4	5	6	7
Alfalfa, lb.	36	10	10	10	10	10	10
Barley, lb.	64	44.4	44.4	26.9	26.9	19.7	19.7
Lowland Hay, lb.		45.0	45.0	60.0	60.0	60.0	60.0
Soybean Oil Meal, lb.				2.0	2.0	4.2	4.2
Stabilized Animal Fat, lb.						5.0	5.0
Cobaltized-iodized Salt, lb.	_ad lib_.	0.6	0.6	0.6	0.6	0.6	0.6
Bone Meal, lb.				0.5	0.5	0.5	0.5
Total, lb.	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Dry Matter, %	90.7	91.8	92.3	92.4	92.1	91.5	88.0
Crude Protein, %	13.3	12.7	13.5	13.2	13.9	13.3	13.2
Gross Energy, Therms/lb.	1.96	2.07	2.02	2.07	2.00	2.20	2.20

Soybean oil meal was added to the rations for Lots 4 to 7, to maintain the level of protein approximately constant in all rations; bone meal was added to increase phosphorus levels above minimum requirements. Additions of stabilized fat were made to rations for Lots 6 and 7 to increase their energy content.

Lambs were marketed as they reached a weight of 95 to 105 pounds. The experiment was terminated after 75 days and the remaining lambs marketed regardless of weight or finish.

Results and Discussion

The results of the trial are summarized in Table 3. Analyses of variance of rate of gain and adjusted rate of gain, and significant mean differences are presented in Appendix A, Tables I and II.

There was no appreciable difference in performance of lambs fed non-pelleted rations (Lots 1, 2, 4 and 6). No significant differences were found in rate of gain, and there was little difference in feed consumed per day or per unit of gain. The average daily gross energy consumption of lambs in Lots 2, 4 and 6 was 12 per cent higher, and their gross energy requirement per unit gain was nine per cent greater than for the lambs in Lot 1, fed the alfalfa-barley ration.

Pelleting of the rations for lambs resulted in greater feed intake, increased rate of gain and improved efficiency of feed utilization. On the average, lambs fed pelleted rations (Lots 3, 5 and 7) consumed 35 per cent more feed daily, gained 44 per cent faster and required seven per cent less feed and 10 per cent less gross energy per unit of gain than those fed similar non-pelleted rations (Lots 2, 4 and 6).

Levels of lowland hay in the ration had no effect on rate of gain of lambs fed the pelleted rations, but the lambs receiving the pelleted rations containing 60 per cent of lowland hay consumed more feed than those fed the pelleted 45 per cent lowland hay ration, and, as a consequence, their efficiency of feed utilization was somewhat lower. Pelleting improved feed and energy conversion 15 and 14 per cent respectively on the 45 per cent lowland hay ration (Lot 3 vs. Lot 2), but only 0.3 and 4 per cent respectively on the 60 per cent lowland hay ration (Lot 5 vs. Lot 4).

Table 3. Effects of Pelleting, Level of Lowland Hay and Supplemental Fat on Rate of Gain and Efficiency of Feed Utilization of Lambs

	Alfalfa -Barley	45% Lowland Hay		60% Lowland Hay		5% Animal Fat, 60% Lowland Hay	
		Chopped	Pelleted	Chopped	Pelleted	Chopped	Pelleted
Lot No.	1	2	3	4	5	6	7
No. of Lambs	8	8	8	8	8	8	8
Av. No. Days Fed	66.8	64.4	50.9	62.0	56.1	68.5	52.0
Av. Initial Wt., lb.	70.3	70.4	70.5	70.6	70.5	70.5	70.9
Av. Total Gain, lb.	23.3	24.4	26.7	22.0	28.6	23.3	26.7
Av. Daily Gain, lb.	0.35	0.38	0.53	0.35	0.51	0.34	0.51
Adjusted Daily Gain, lb.	0.41	0.41	0.48	0.39	0.38	0.42	0.43
Feed/Day, lb.	2.98	3.22	3.81	3.09	4.43	2.85	4.10
Crude Protein /Day, lb.	0.36	0.38	0.48	0.38	0.57	0.35	0.48
Gross Energy /Day, Therms	5.29	6.13	7.11	5.92	8.16	5.74	7.94
Feed/100 lb. Gain, lb.	854	851	725	871	868	842	798
Gross Energy/100 lb. Gain, Therms	1517	1617	1351	1666	1598	1694	1544
Market Grade:							
Choice and Good	6	7	8	2	7	7	7
Medium and Fair	2	1		6	1	1	1

The results for Lots 6 and 7 vs. Lots 4 and 5 indicate that addition of animal fat to the 60 per cent lowland hay rations reduced daily feed consumption but did not affect rate of gain. Although the rations containing animal fat were higher in gross energy than the unsupplemented rations, consumption of gross energy by lambs was slightly lower because of the reduction in feed intake. There were no appreciable differences in efficiency of utilization of feed or gross energy. This effect is similar to that reported by Perry _et al_. (1959) and Wise _et al_. (1959) who noted that addition of animal fat to a ration resulted in reduced feed intake.

In this trial, average daily gain was directly related to feed intake. A highly significant correlation ($r_{xy} = 0.88$; $P < 0.01$) was found between average daily gain and average feed consumed. When average daily gain was adjusted for feed intake (Table 3), no significant differences were found in rate of gain between lots. This indicates that increased rate of gain of lambs fed pelleted rations was largely the result of increased feed intake. Similar results have been reported by Meyer _et al_. (1959b).

Lambs fed pelleted rations reached market weight and finish earlier than those fed non-pelleted rations. At the end of the trial, only two of the lambs fed pelleted rations, but ten of those fed non-pelleted rations, had failed to attain satisfactory market weight and finish.

Summary

(1) Native lowland hay was fed to lambs at levels of 45 and 60 per cent of the ration with results comparable to those obtained by feeding alfalfa and barley.

(2) Pelleting of rations containing lowland hay resulted in a marked increase in feed intake and rate of gain. When compared to similar non-pelleted rations, feed and energy conversion were improved on the 45 per cent lowland hay ration, but were not affected on the 60 per cent lowland hay ration.

(3) Increasing the energy content of the 60 per cent lowland hay rations by the addition of stabilized animal fat reduced feed intake but had no effect on rate of gain or energy required per unit of gain.

Trial 2

Object

The results of the previous trial indicated that pelleting of rations containing 45 and 60 per cent of native lowland hay resulted in increased feed intake, faster rate of gain and improved feed efficiency. Since these effects may indicate differences in digestion, a trial was undertaken to study the effects of pelleting of rations containing 45, 60 and 90 per cent of lowland hay on rate of gain and efficiency of feed utilization by lambs and on apparent digestibility of the ration.

Experimental

Twenty-eight wether lambs (Suffolk and crossbred Hampshire, Suffolk and Romnelet) from the University of Alberta flock were divided into seven groups of four lambs each. The lambs were placed in individual pens and hand-fed all they would consume twice a day.

The formulation of the rations fed is shown in Table 4.

Apparent digestion coefficients of dry matter, crude protein and gross energy were determined by the total fecal collection technique. The wethers were fed the experimental rations for at least three weeks before

fecal collections commenced. Total feces from each wether were collected daily by means of canvas bags (Laverty, 1961). Polyethylene sacks (18" x 24") were used to line the inside of each canvas bag and were replaced daily during the seven-day collection period. Daily fecal excretions from each wether were mixed and dried for 48 hours at 80° C. in a mechanical convection oven. A representative portion of the dried feces from each wether was ground, composited and a sample retained for chemical analyses. The feed supplied and unconsumed feed were also sampled for chemical analyses. Feed and fecal samples were ground in a Wiley No. 1 Mill and stored in closed glass jars until analyzed.

Table 4. Rations for Trial 2

Lot Number		1	2	3	4	5	6	7
Alfalfa,	lb.	33.0	10.0	10.0	10.0	10.0	10.0	10.0
Barley,	lb.	67.0	41.2	41.2	28.2	28.2		
Lowland Hay,	lb.		45.0	45.0	60.0	60.0	90.0	90.0
Soybean Oil Meal,	lb.		3.2	3.2	1.2	1.2		
Cobaltized-iodized Salt*,	lb.		0.6	0.6	0.6	0.6		
Total,	lb.	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Dry Matter,	%	91.5	92.7	93.4	92.8	93.1	93.2	93.8
Crude Protein,	%	15.0	14.1	15.2	12.7	14.0	12.1	12.4
Gross Energy,	Therms/lb.	1.94	1.93	1.94	1.92	1.94	1.94	1.96

* Mineral mix of 50% salt and 50% bone meal available free choice to all lambs.

Three of the lambs in Lot 1 and all of the lambs in Lot 3 were marketed after 61 days on test; these lambs had reached weights of 90 pounds

or over. The heaviest lamb in each of Lots 2, 4 and 5 was also marketed at this time for carcass and rumen comparisons. All of the lambs in Lot 6 and two in Lot 7 were marketed because of weight losses and debility. The remaining lambs were marketed after 85 days on test.

Photographs of the rumen epithelium were taken at time of slaughter.

Results and Discussion

A summary of the results obtained is presented in Table 5. Statistical analyses of the data indicated that treatments had highly significant effects on dry matter consumed daily and on digestion coefficients for dry matter, crude protein and gross energy (Appendix A, Table I).

The level of lowland hay in the rations affected rate of gain of the lambs. Low rates of gain or loss of weight were noted in the groups fed non-pelleted rations containing lowland hay (Lots 2, 4 and 6), with rate of gain decreasing as level of lowland hay in the ration increased.

Daily consumption of dry matter decreased as the level of lowland hay in the ration was increased. Dry matter intake of lambs fed 90 per cent lowland hay (Lot 6) was significantly lower (Appendix A, Table II) than that of lambs fed 45 or 60 per cent lowland hay (Lots 2 and 4). The low feed intake noted in the trial may have been caused by the presence of wild barley in the lowland hay.

Pelleting of the rations resulted in marked increases in feed intake, rate of gain and efficiency of feed utilization (Lots 3, 5 and 7 vs. Lots 2, 4 and 6 respectively). In the case of lambs fed the pelleted ration containing 45 per cent of lowland hay (Lot 3), feed intake and rate of gain were equivalent to those of lambs fed the alfalfa-barley ration (Lot 1). As the level of lowland hay in the ration increased, feed intake and rate of gain declined (Lots 5 and 7), and the amount of feed and digested energy required per unit of gain increased.

Table 5. Effects of Pelleting and Level of Lowland Hay on Rate of Gain and Efficiency of Feed Utilization of Lambs and on Apparent Digestibility of the Ration

	Alfalfa -Barley	45% Lowland Hay		60% Lowland Hay		90% Lowland Hay	
		Chopped	Pelleted	Chopped	Pelleted	Chopped	Pelleted
Lot No.	1	2	3	4	5	6*	7**
Av. No. Days Fed	66	77	61	77	77	54	61
Av. Initial Wt., lb.	63.2	63.0	63.2	63.5	63.2	62.8	63.0
Av. Total Gain, lb.	39.0	16.5	38.0	4.3	35.5	-13.3	5.5
Av. Daily Gain, lb.	0.59	0.21	0.62	0.06	0.46		0.09
Feed/Day, lb.	3.32	1.94	3.62	1.51	3.54	0.64	2.59
Dry Matter/Day, lb.	3.03	1.79	3.39	1.39	3.30	0.59	2.43
Digestibility Dry Matter, %	78	72	65	71	70	50	58
Digested Dry Matter/Day, lb.	2.36	1.29	2.20	0.99	2.31	0.30	1.41
Digestibility Crude Protein, %	73	70	70	71	75	57	63
Digested Crude Protein/Day, lb.	0.33	0.18	0.36	0.13	0.34	0.04	0.19
Digestibility Gross Energy, %	76	71	64	70	69	57	58
Digested Gross Energy/Day, Therms	4.47	2.45	4.21	1.87	4.42	0.65	2.76
Feed/100 lb. Gain, lb.	563	902	582	2735	765		2882
Digested Energy /100 lb. Gain, Therms	759	1145	675	3411	953		3073
Market Grade:							
Choice and Good	4		3		4		
Medium and Fair		4		3			
Cull and Condemned			1'			3	3

*One lamb died after 34 days on test.
**One lamb died of impaction after 41 days on test.
'Carcass condemned because of pneumonia.

The digestion studies that were conducted in conjuction with the feeding trial yielded some interesting data, which are also summarized in Table 5. Digestion coefficients obtained for dry matter, crude protein and gross energy were very similar in lots receiving non-pelleted rations containing 45 and 60 per cent of lowland hay; digestibility of the nutrients varied between 70 and 72 per cent. These were somewhat lower than corresponding coefficients obtained for the alfalfa-barley ration (Lot 1), in which digestibility varied between 73 and 78 per cent. In the non-pelleted ration containing 90 per cent of lowland hay, digestion coefficients were significantly lower than in the other rations (Appendix A, Table II).

The effects of pelleting on digestibility varied depending upon the level of lowland hay in the ration. Pelleting resulted in a significant decrease in digestibility of dry matter and gross energy with the 45 per cent lowland hay ration, had no effect on any of the digestion coefficients with the 60 per cent lowland hay ration, and increased digestibility of dry matter and crude protein with the 90 per cent lowland hay ration. In spite of the increase in digestibility noted when the 90 per cent lowland hay ration was pelleted, digestion coefficients were still significantly lower than with rations with 45 or 60 per cent of lowland hay.

Lambs fed the alfalfa-barley ration (Lot 1) and pelleted rations containing 45 and 60 per cent of lowland hay (Lots 3 and 5) attained satisfactory market weight and finish; lambs fed the other rations did not.

Rations containing lowland hay had marked effects on color of the rumen epithelium and on papillary development (Fig. 1). Rumina from lambs fed alfalfa and barley (Lot 1) appeared healthy and had large, well-developed papillae. The inclusion of lowland hay in the ration resulted in a dark discoloration of the rumen epithelium; papillary development decreased with increasing levels of lowland hay in the ration. Pelleting

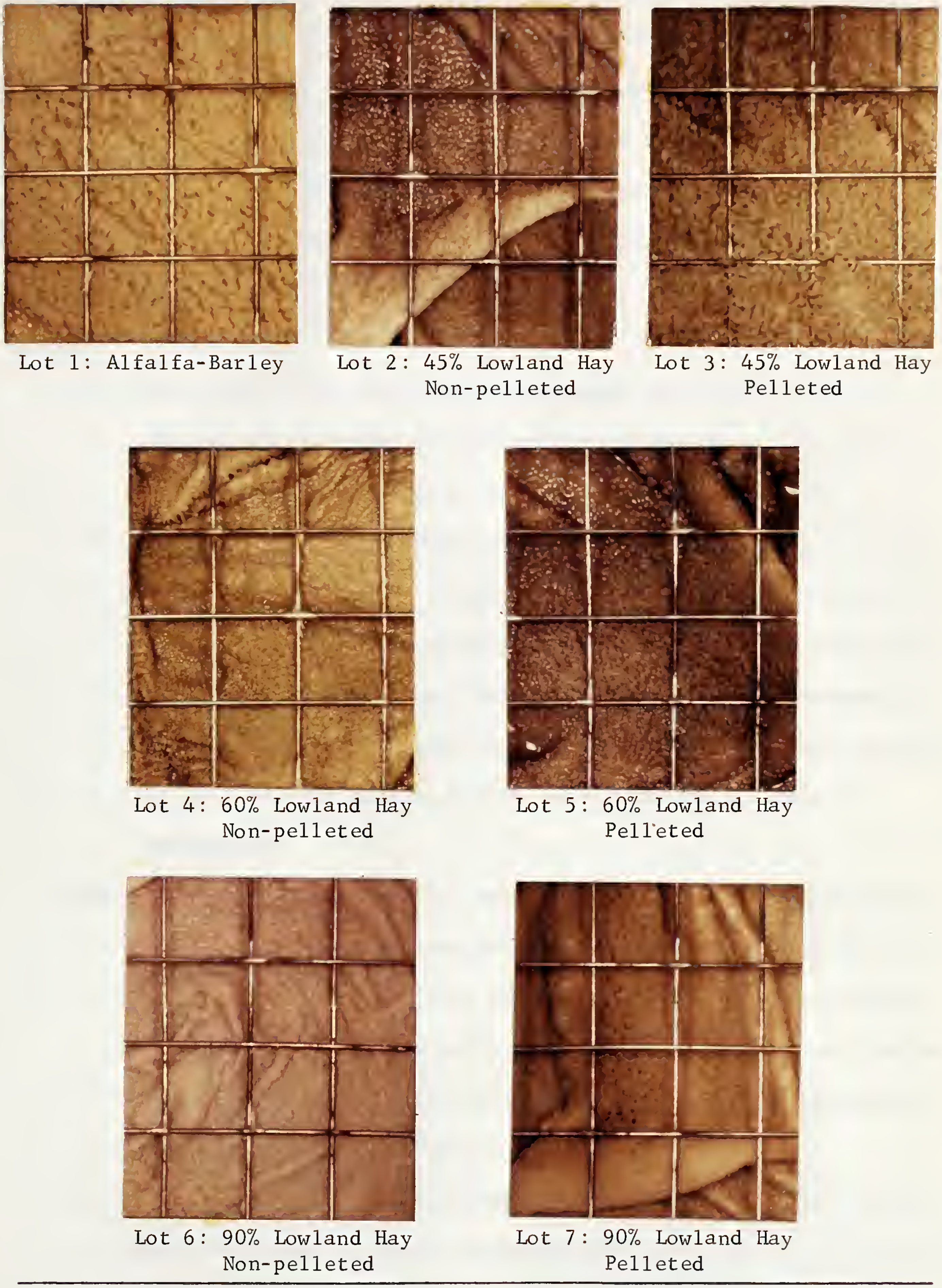


Figure 1. Effects of Pelleting and Level of Lowland Hay on the Rumen Epithelium.

of the rations appeared to increase papillary development as compared to similar non-pelleted rations. A reddish discoloration, suggesting inflammation, was noted in the epithelium of rumina from lambs fed 90 per cent lowland hay rations; this was also evident in the rumen of one lamb fed the pelleted 45 per cent lowland hay ration. It is possible that the inflammation may have influenced feed intake and possibly efficiency of feed utilization.

Summary

(1) Non-pelleted rations containing lowland hay were consumed in low amounts by lambs and produced unsatisfactory rates of gain, inefficient utilization of feed and low carcass quality.

(2) Pelleting the rations resulted in marked improvements in rate of gain and efficiency of feed utilization of lambs. Rate of gain and efficiency of feed utilization by lambs fed pelleted rations containing 45 per cent of lowland hay were comparable to those of lambs fed alfalfa and barley; however, feed required per unit of gain increased with each increase in level of lowland hay.

(3) Digestion coefficients for dry matter, crude protein and gross energy were similar in non-pelleted rations containing 45 and 60 per cent of lowland hay, but were lower than those obtained with the alfalfa-barley ration. Increasing the level of lowland hay to 90 per cent of the ration caused significant decreases in all digestion coefficients determined.

(4) Pelleting resulted in decreased digestibility of dry matter and gross energy in the ration containing 45 per cent of lowland hay, had no effect on digestion coefficients in the ration containing 60 per cent of lowland hay, and increased digestibility of dry

matter and crude protein in the ration containing 90 per cent of lowland hay.

(5) Rations containing lowland hay resulted in a dark discoloration and reduced papillary development in the rumen epithelium. Pelleting the rations appeared to increase papillary development, as compared to similar non-pelleted rations. Very high levels of lowland hay in the ration appeared to produce an inflammation of the rumen epithelium.

Trial 3

Object

Since rate of growth and feed efficiency in the previous trial were very low when 90 per cent of lowland hay was included in the ration, a trial was conducted to study the effects of increased nitrogen from supplemental urea and amino acids on performance of lambs fed pelleted and non-pelleted rations containing high levels of native lowland hay.

Experimental

Forty-two ewe lambs of range type (mixed Rambouillet and Romnelet breeding) were divided into seven groups of six lambs each and fed the experimental rations shown in Table 6. Except for the controls (Lot 1), all lambs were self-fed.

Molasses was added to the rations containing lowland hay in an attempt to improve palatability and feed intake. It was also considered that molasses would serve as a source of readily available energy to promote nitrogen utilization by rumen micro-organisms in lambs fed rations containing urea, as suggested by Coombe et al. (1960). In the non-pelleted rations, the molasses, diluted with warm water, and containing urea and amino acids as required, was sprayed over the hay from a watering can. This was followed by thorough mixing.

Table 6. Rations for Trial 3

Lot Number		1	2	3	4	5	6	7
Alfalfa,	lb.	42.0	10.0	10.0	10.0	10.0	10.0	10.0
Barley,	lb.	58.0						
Lowland Hay,	lb.		84.0	84.0	83.0	83.0	82.8	82.8
Molasses,	lb.		5.0	5.0	5.0	5.0	5.0	5.0
Urea,	lb.				1.0	1.0	1.0	1.0
L-lysine,	lb.						0.1	0.1
DL-methionine,	lb.						0.1	0.1
Salt & Mineral*,	lb.	*ad lib*.	1.0	1.0	1.0	1.0	1.0	1.0
Total,	lb.	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Dry Matter,	%	91.7	93.2	93.9	92.6	93.0	92.6	93.1
Crude Protein,	%	14.4	10.3	11.6	13.3	14.0	13.8	14.4
Gross Energy,	Therms/lb.	1.97	1.96	1.99	2.00	1.98	1.98	1.95

*50% Cobaltized-iodized Salt and 50% Bone Meal.

All of the lambs in Lot 3, and the three heaviest lambs in each of the other lots, were marketed after six weeks on test. The remainder were marketed two weeks later, when the trial was terminated.

Results and Discussion

The results of the trial are summarized in Table 7. Analyses of variance of the data and significant mean differences are presented in Appendix A, Tables I and II.

No differences were noted in rate of gain of lambs self-fed non-pelleted rations containing lowland hay (Lots 2, 4 and 6). The lambs consumed approximately the same amount of feed daily, and required similar

Table 7. Effects of Pelleting and Nitrogen Supplements on Rate of Gain and Efficiency of Feed Utilization of Lambs

	Alfalfa -Barley	Non-Supplemented		Urea		Urea + Amino Acids	
		Chopped	Pelleted	Chopped	Pelleted	Chopped	Pelleted
Lot No.	1	2	3	4	5	6	7
No. of Lambs	6	6	6	6	6	6	6
Av. No. Days Fed	49	49	42	49	49	49	49
Av. Initial Wt., lb.	82.5	83.5	82.5	82.8	86.3	83.0	82.3
Av. Total Gain, lb.	13.7	8.0	19.2	8.3	20.7	8.2	16.8
Av. Daily Gain, lb.	0.28	0.16	0.46	0.17	0.42	0.17	0.34
Adjusted Daily Gain, lb.	0.40	0.22	0.30	0.20	0.35	0.23	0.28
Feed/Day, lb.	2.78	3.15	4.52	3.35	3.99	3.12	3.90
Crude Protein /Day, lb.	0.37	0.30	0.49	0.41	0.52	0.40	0.52
Gross Energy /Day, Therms	5.02	5.76	8.44	6.20	7.34	5.74	7.08
Feed/100 lb. Gain, lb.	999	1930	990	1970	950	1880	1140
Gross Energy/100 lb. Gain, Therms	1806	3526	1851	3648	1750	3447	2069
Market Grade:							
Choice and Good	5	3	6	4	5	3	5
Medium and Fair	1	3		2	1	3	1

amounts of feed and gross energy per unit of gain, indicating that nitrogen supplements did not improve utilization of the rations. Although lambs fed non-pelleted lowland hay rations consumed 16 per cent more feed daily than lambs fed alfalfa and barley (Lot 1), rate of gain was 39 per cent lower (approached significance) and 93 per cent more feed and 96 per cent more gross energy were required per unit of gain.

The results of the trial again indicated that pelleting of the rations resulted in a marked increase in rate of gain and efficiency of feed utilization. Average daily gain was increased 144 per cent, feed intake increased 28 per cent and feed required per unit of gain declined 47 per cent, as compared to results with the non-pelleted rations containing lowland hay. Lambs fed pelleted rations consumed 48 per cent more feed daily and gained at a faster rate than lambs fed alfalfa and barley (difference in rate of gain between lambs in Lots 7 and 1 only approached significance). However, they required approximately the same amount of feed and gross energy per unit of gain.

The addition of nitrogen supplements may have affected rate of gain and feed intake of lambs fed pelleted rations. Rate of gain of lambs decreased (non-significant) when urea was added to the ration (Lot 5 vs. Lot 3) and decreased still further (approached significance) when both urea and amino acids were added (Lot 7 vs. Lot 3). The addition of urea and of urea and amino acids to pelleted rations reduced feed intake by lambs 12 and 14 per cent respectively, compared to that of lambs fed the unsupplemented pelleted ration. The addition of urea alone resulted in a slight improvement in utilization of feed and gross energy.

It is evident that supplements of lysine and methionine did not produce a beneficial effect on performance of lambs fed rations containing urea. While in some reports it has been noted that supplements of lysine

or methionine have improved performance of lambs and steers fed rations containing urea, (Gossett *et al*., 1960; Noble *et al*., 1955; Barth *et al*., 1959), other reports have indicated no beneficial effects from amino acid supplements (Harbers *et al*., 1961; Gossett *et al*., 1961).

Rate of gain was found to be associated with feed intake ($r_{xy} = 0.82$; $P < 0.05$). When rate of gain was adjusted for average feed intake, treatments were found to have significant effects (Appendix A, Table I). The adjusted daily gain of lambs fed alfalfa and barley (Lot 1) was significantly higher than that of lambs fed non-pelleted rations containing lowland hay (Lots 2, 4 and 6). This might be expected, as it was found in the previous trial that digestibility was low in rations containing a very high level of lowland hay. The adjusted gain of lambs fed pelleted rations was intermediate between that of lambs fed the alfalfa-barley ration and lambs fed non-pelleted rations containing lowland hay. This may have resulted from increased digestibility of dry matter and crude protein as a result of pelleting rations containing high levels of lowland hay, as observed in the previous trial. Pelleting of the ration containing supplemental urea (Lot 5 vs. Lot 4) resulted in significantly increased adjusted rate of gain, possibly reflecting increased utilization of gross energy.

Lambs fed pelleted rations reached market weight and finish earlier than other lambs in the trial. Nine lambs fed non-pelleted rations, but only two lambs fed pelleted rations failed to reach satisfactory weight and finish by the end of the trial.

<u>Summary</u>

(1) Lambs fed non-pelleted rations containing 84 per cent of lowland hay consumed more feed, but gained less weight than lambs fed alfalfa and barley.

(2) Pelleted rations resulted in marked increases in rate of gain, feed intake and efficiency of feed utilization as compared to similar non-pelleted rations.

(3) Supplementation of rations containing lowland hay with urea and with urea and amino acids did not improve rate of gain of fattening lambs. The addition of urea alone in the pelleted ration may have improved efficiency of utilization of feed and gross energy.

Trial 4

Object

The trial was conducted to obtain further data on the effects of pelleting of rations containing high levels of lowland hay on apparent digestibility of dry matter, crude protein and gross energy.

Experimental

Five mature wethers (2 Hampshire and 3 Suffolk) were prepared with Jarrett (1948) fistulae, penned together and fed individually twice a day.

The rations were similar to those fed in Lots 1, 4, 5, 6 and 7 of Trial 2 (Table 4). Salt was omitted from rations for Lots 4 and 5; barley was increased correspondingly. A salt-mineral mix was available free choice to all wethers. Results of chemical analyses of the rations are shown in Table 8.

Table 8. Dry Matter, Crude Protein and Gross Energy Content of the Rations

Lot Number		1	4	5	6	7
Dry Matter,	%	88.9	90.2	92.0	90.5	92.6
Crude Protein,	%	13.3	14.0	14.1	12.3	14.9
Gross Energy,	Therms/lb.	2.00	2.04	2.01	2.02	1.99

The wethers were fed the alfalfa-barley ration for three weeks prior to commencement of the trial (pretreatment). During the trial, each of the lowland hay rations was fed to one wether for four weeks (Lots 4 to 7). The rations fed to wethers in Lots 4 and 5, and in Lots 6 and 7, were reversed, so that each wether received the same ration, but in a different physical form, for another four weeks. One wether was continued on the control ration during the entire trial period. The trial was terminated after eight weeks.

Digestibility studies, as described in Trial 2, were conducted throughout the fourth and eighth weeks; the values reported represent the average of two determinations on each ration.

Results and Discussion

Digestion coefficients obtained for dry matter, crude protein and gross energy are shown in Table 9.

Table 9. Effects of Pelleting and Level of Lowland Hay on Apparent Digestibility of Dry Matter, Crude Protein and Gross Energy

		DIGESTION COEFFICIENTS (per cent)		
Lot No.	Ration	Dry Matter	Crude Protein	Gross Energy
1	Control, Alfalfa-Barley	74	74	75
4	60% Lowland Hay, Chopped	62	69	64
5	" " ", Pelleted	64	68	64
6	90% Lowland Hay, Chopped	54	64	57
7	" " ", Pelleted	56	67	56

Lower digestion coefficients were obtained with rations containing lowland hay than with the alfalfa-barley ration; the greatest decrease was obtained with the 90 per cent lowland hay ration. Digestibility of

crude protein was not reduced as much as that of dry matter and gross energy. Pelleting the rations had no appreciable effects on digestion coefficients.

In general, the results agree with those obtained in Trial 2; digestibility decreased with increasing levels of lowland hay and was very low in rations containing 90 per cent of lowland hay. However, pelleting of the ration containing 90 per cent of lowland hay did not result in as great an increase in digestibility of dry matter and crude protein as was noted in the previous trial.

Summary

(1) Digestibility of dry matter, crude protein and gross energy was lower with rations containing lowland hay than with the alfalfa-barley ration; digestibility decreased with increasing levels of lowland hay in the ration.

(2) Pelleting of the rations did not appreciably affect digestion coefficients.

II. Effects of Pelleting and Level of Lowland Hay on Rate of Rumen Fermentation

Status of the Problem

It has been noted that feed intake of ruminants is increased when rations containing high levels of roughage are pelleted. This would seem to imply that grinding and pelleting results in changes in the ration which promote increased rate of digestion in the rumen and increased rate of food passage through the digestive tract, thus allowing greater feed intake. If an increase in rate of digestion does occur, this probably indicates a more active microbial population which should be reflected in the rate of fermentation occurring in the rumen ingesta.

Since information was lacking on the effect of ration variations on rate of fermentation in the rumen, a study was undertaken to assess the effects of pelleting of rations containing varying levels of native lowland hay on rate of rumen fermentation.

Trial 1

Object

To determine the effects of pelleted rations containing varying levels of native lowland hay on rate of rumen fermentation in lambs at varying intervals after feeding.

Experimental

Rate of fermentation, in the rumen contents of three lambs from Lot 6 and four lambs from each of the other lots in the second feeding trial previously outlined, was measured during the sixth to ninth weeks of the trial.

Samples of the rumen contents of two lambs each time, representing comparable pelleted and non-pelleted rations, were obtained immediately before feeding (0 hours) and at 1½, 3 and 6 hour intervals thereafter. At least 24 hours elapsed between successive samples taken from any one lamb. Sampling was done by inserting a stomach tube (lumen diameter 5/16") into the rumen via the mouth and esophagus, using a mouth speculum for swine. The free end of the tube was attached to a side-arm receiving flask which was connected by tubing to a second side-arm 'overflow' flask. A vacuum pump, capable of producing a vacuum of 24 inch Hg., was attached to the overflow flask to withdraw rumen contents into the receiving flask.

Rumen samples were transferred to 250 ml. Erlenmeyer flasks and the flasks were flushed with carbon dioxide and loosely stoppered before transport to the laboratory.

Rate of fermentation was measured by an in vitro procedure adapted from that outlined by Hungate et al. (1955) and modified by Meyer et al. (1959a). Details of the procedure used are outlined in Appendix B. Measurements made included manometric pressures, total gas production and total acid production, each of which may be considered a measure of rate of fermentation.

Results and Discussion

The effects of ration variations and time intervals on rate of in vitro fermentation are depicted graphically in Fig. 2. A linear increase in pressure was noted with all rations at each sampling interval during incubation of rumen samples for one hour. This indicates that there was little change in microbial activity during the period of incubation.

Rate of fermentation was influenced by time of sampling. With rations containing 45 or 60 per cent of lowland hay, rate of fermentation was low just prior to feeding (0 hour), had increased by 1½ hours after feeding and maintained the rapid rate until six hours after feeding. In the control group (alfalfa-barley) rate of fermentation was high at all sampling intervals, while in the groups receiving 90 per cent of lowland hay, rate of fermentation was low.

Level of lowland hay in the ration affected rate of fermentation of rumen contents. With rations containing 45 and 60 per cent of lowland hay, rate of fermentation was lower than with the control ration (alfalfa-barley), but much higher than with the ration containing 90 per cent of lowland hay.

It was evident that pelleting of the rations had no effect on rate of fermentation. Levels of fermentation noted with similar pelleted and non-pelleted rations at each sampling interval were practically the same.

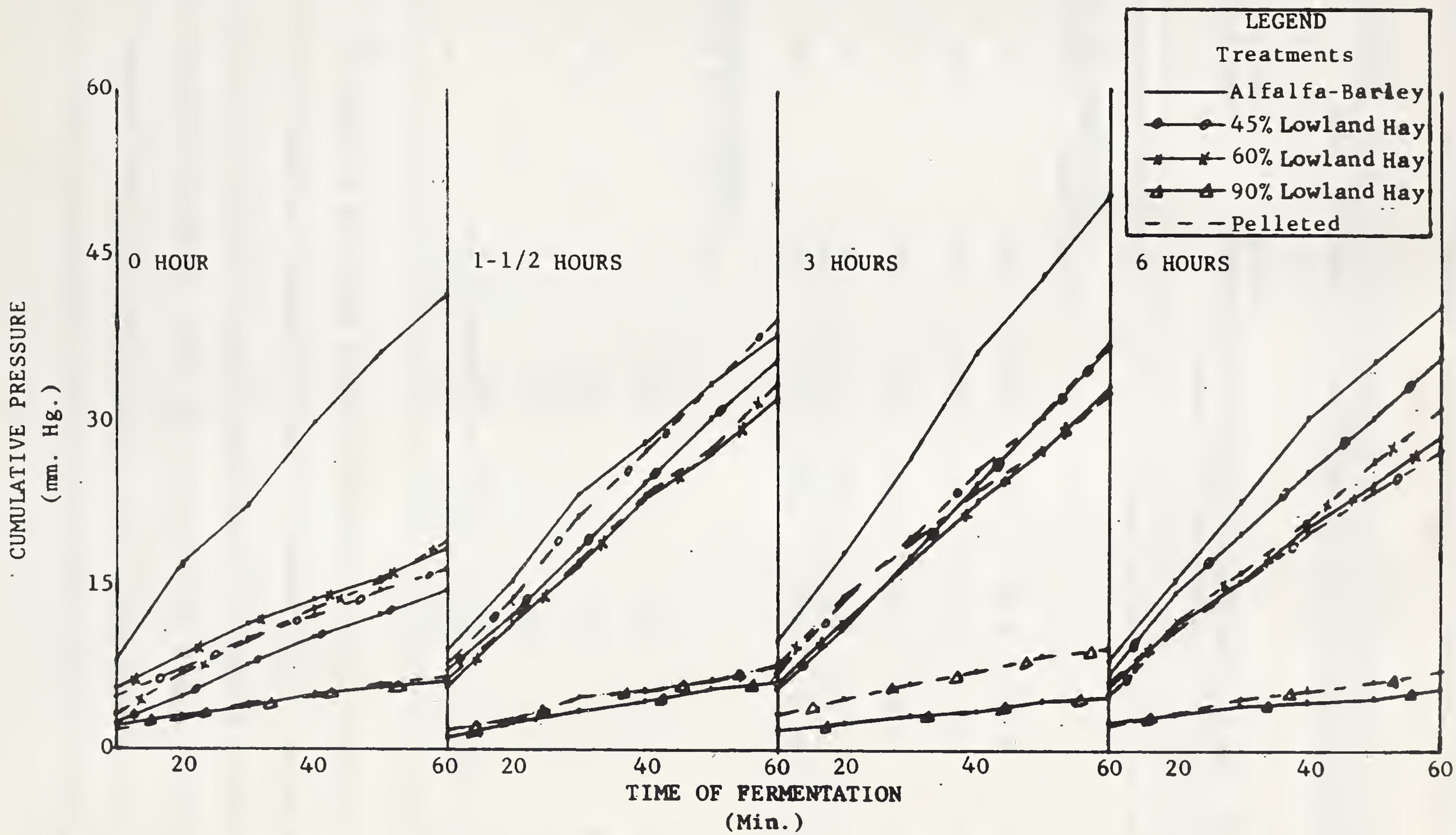


Figure 2. Effects of Pelleting and Level of Lowland Hay on Pressure Produced in Manometric Fermentations: Trial 1

The production of total gas in manometric fermentations of rumen samples is summarized in Table 10. Analysis of variance of the data and significant mean differences are presented in Appendix C, Tables I and II.

Table 10. Effects of Pelleting and Level of Lowland Hay on Production of Total Gas and Acid in Manometric Fermentations: Trial 1

Hours After Feeding Lambs	Alfalfa -Barley	45% Lowland Hay Chopped	45% Lowland Hay Pelleted	60% Lowland Hay Chopped	60% Lowland Hay Pelleted	90% Lowland Hay Chopped	90% Lowland Hay Pelleted
Lot No.	1	2	3	4	5	6	7
Total Gas Production (ml./100 ml. rumen liquid)							
0	82.2	28.9	32.6	36.7	37.6	13.0	13.6
1½	74.4	67.8	77.4	61.3	64.5	12.5	14.6
3	96.0	70.5	71.4	62.3	62.8	9.5	18.4
6	79.8	71.2	54.5	56.5	60.2	11.1	14.2
Av.	83.1	59.6	59.0	54.2	56.3	11.5	15.2
Total Acid Production (mM./100 ml. rumen liquid)							
0	1.79	0.50	0.63	0.62	0.75	0.21	0.16
1½	1.45	1.62	1.62	1.30	1.27	0.13	0.28
3	2.06	1.32	1.46	1.37	1.63	0.12	0.44
6	1.61	1.24	1.01	1.02	1.16	0.29	0.25
Av.	1.73	1.17	1.18	1.08	1.20	0.18	0.28

The results were very similar to those noted above when increase in pressure was used as a measure of rate of fermentation. The production of gas was significantly lower in samples taken prior to feeding (0 hour) than in those taken after feeding. This was the result of a much lower production of gas in samples obtained prior to feeding from lambs in Lots 2 to 5 (45 and 60 per cent lowland hay rations) than in those obtained after feeding these lambs.

A summary of the data on acid production in manometric fermentations is shown in Table 10, and illustrated graphically in Fig. 3. Analysis of variance of the data and significant mean differences are shown in Appendix C, Tables I and II.

The results were similar to those obtained for increase in pressure and for total gas production. Similar amounts of acid were produced in rumen samples from lambs fed non-pelleted rations containing 45 and 60 per cent of lowland hay (Lots 2 and 4), but these were 35 per cent lower than that produced in samples from lambs fed the control ration. Acid production in samples from lambs fed the non-pelleted ration containing 90 per cent of lowland hay (Lot 6) was significantly less than that produced in samples from lambs fed the other non-pelleted rations. Only 10 per cent as much acid was produced as in samples from lambs fed the control ration (Lot 1).

It was evident that pelleting of the rations did not have marked effects on acid production. The amount of acid produced in samples from lambs fed pelleted rations was slightly higher than in those from lambs fed non-pelleted rations, but the differences were not found to be significant. Although there was a large percentage increase in acid production when the 90 per cent lowland hay ration was pelleted, the molar increase was very small (0.10 mM/100 ml. rumen liquid) and non-significant.

The results obtained in the manometric fermentations reflect a high rate of fermentation in rumen contents from lambs fed alfalfa and barley, a lower rate from rations containing 45 and 60 per cent of lowland hay, and a very low rate in rumen contents from lambs fed rations containing 90 per cent of lowland hay. Pelleting the rations had no appreciable effects on rate of fermentation and could not account for the effects noted on feed intake and rate of gain as a result of pelleting.

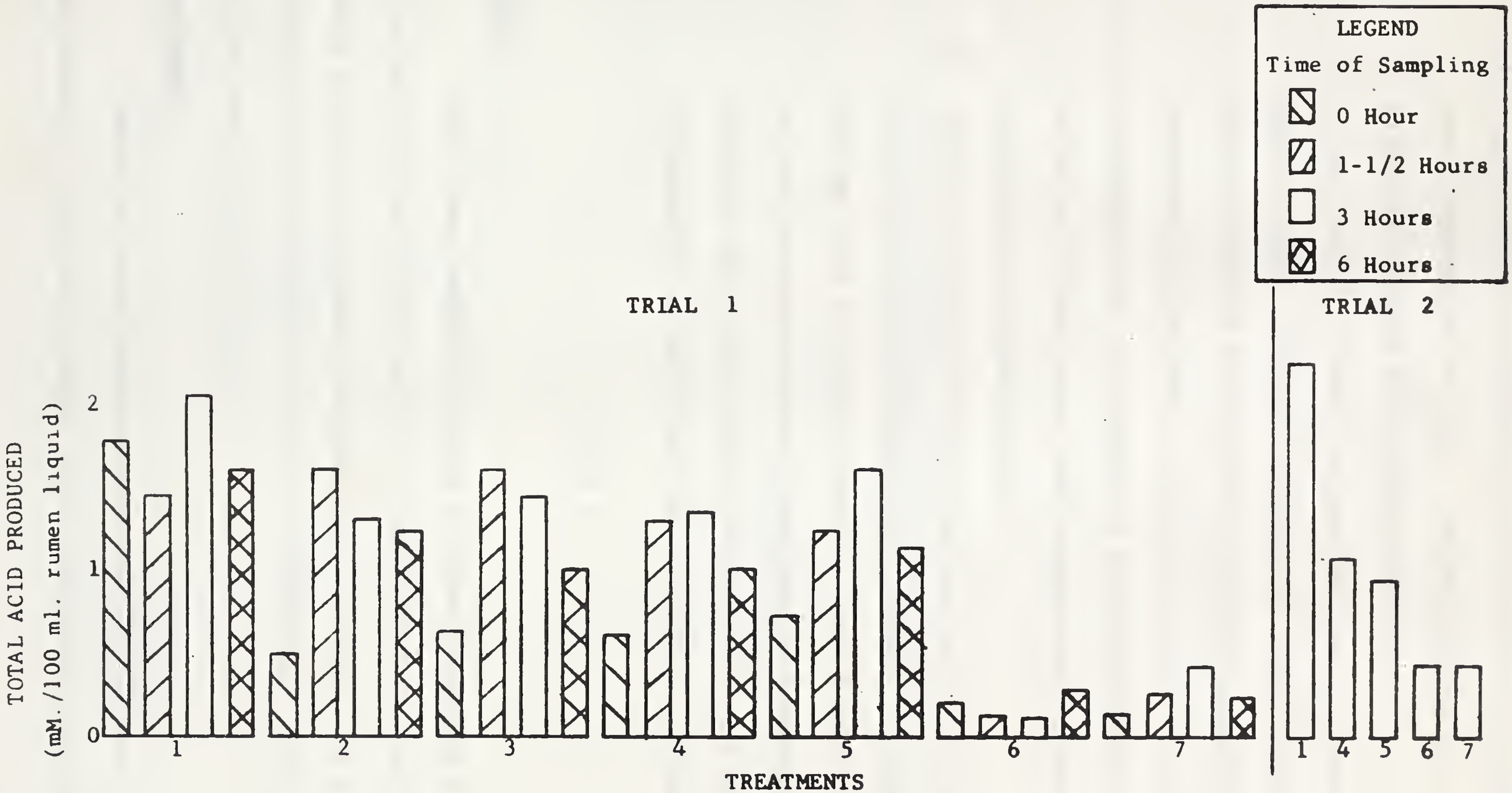


Figure 3. Effects of Pelleting and Level of Lowland Hay on Total Acid Produced in Manometric Fermentations

Summary

(1) A steady rate of fermentation was obtained in incubation of rumen samples for one hour, indicating there was no change in microbial activity during this period.

(2) A high rate of fermentation was obtained with the alfalfa-barley ration. Rations containing 45 and 60 per cent of lowland hay reduced fermentation rates similar amounts, and rations containing 90 per cent of lowland hay resulted in very low rates of fermentation, as compared to the alfalfa-barley ration.

(3) Pelleting of the rations had no appreciable effects on rate of fermentation of rumen samples.

(4) Rations containing 45 and 60 per cent of lowland hay resulted in a lower rate of fermentation in samples taken just before feeding, than in samples taken $1\frac{1}{2}$, 3 and 6 hours after feeding. At each sampling interval a high rate of fermentation was obtained on the alfalfa-barley ration, and a very low rate of fermentation on the 90 per cent lowland hay rations.

Trial 2

Object

Since rumen samples obtained by stomach tube may be diluted with saliva and yield unreliable results in fermentation studies, it was considered desirable to obtain additional data for comparison with those of the previous trial, by measuring rate of fermentation in rumen samples obtained from fistulated wethers.

Experimental

Samples of rumen contents were obtained from each of five fistulated wethers in the fourth feeding trial (previously outlined on p. 36)

in the third week of the pretreatment period and in the fourth week of each of the two replicates of the trial. All samples were taken three hours after the morning meal.

Polyethylene tubing (lumen diameter ½") was inserted into the rumen through the fistula. A large rubber bulb was used to draw rumen contents into the tube for transfer to a 250 ml. Erlenmeyer flask. After collection of the samples, the procedure followed was the same as that outlined in the previous trial.

Results and Discussion

Production of total gas in manometric fermentations of rumen samples is summarized in Table 11.

Table 11. Effects of Pelleting and Level of Lowland Hay on Production of Total Gas and Acid in Manometric Fermentations: Trial 2

Hours After Feeding Wethers	Pre-treatment	Alfalfa -Barley	60% Lowland Hay		90% Lowland Hay	
			Chopped	Pelleted	Chopped	Pelleted
Lot No.		1	4	5	6	7
Total Gas Produced (ml./100ml. rumen liquid)						
3	119.5	79.6	32.3	33.2	12.6	10.8
Total Acid Produced (mM./100 ml. rumen liquid)						
3	2.78	2.27	1.08	0.96	0.44	0.44

Rate of fermentation in rumen samples was affected by the level of lowland hay in the ration. Gas production in samples from wethers fed the alfalfa-barley ration (Lot 1) was significantly higher than in those from wethers fed rations containing lowland hay (Appendix C, Table II). More than twice as much gas was produced in samples from wethers fed rations containing 60 per cent of lowland hay than in those from wethers fed rations

containing 90 per cent of lowland hay. The difference was not found to be significant, probably because of the small number of animals and determinations involved in the comparison. Gas production in rumen samples from wethers fed the alfalfa-barley ration was lower during the trial than in the pretreatment period; no explanation for this reduction can be offered.

Pelleting of the rations had no effect on production of total gas in the rumen samples; similar amounts of gas were produced in samples from wethers fed pelleted and non-pelleted rations containing the same level of lowland hay.

Data on acid production in manometric fermentations of rumen samples are summarized in Table 11 and illustrated graphically in Fig. 3.

The results were similar to those noted above for gas production. The greatest amount of acid was found in samples from wethers fed alfalfa and barley (Lot 1), and each level of lowland hay in the ration (60 and 90 per cent) resulted in successive decreases in the amount of acid produced. Significant differences were not detected (Appendix C, Table I), again probably because of the small number of determinations involved. The amount of total acid produced was not affected by pelleting of the rations.

The results agree with those obtained in Trial 1, suggesting a high rate of fermentation in rumen contents of sheep fed alfalfa and barley, a lower fermentation rate on 60 per cent lowland hay rations, and a very low rate of fermentation on 90 per cent lowland hay rations. Comparison of pressure increase and of acid production in fermentation of comparable samples by a t-test revealed no significant differences between trials (Appendix C, Table III).

The lack of effect of pelleting on fermentation rates indicates that differences in rate of fermentation in the rumen do not explain the increased consumption that occurs when pelleted rations are fed.

Summary

(1) Rumen samples from wethers fed alfalfa and barley gave greater production of total gas and total acid than similar samples from wethers fed lowland hay rations, indicating a faster rate of rumen fermentation.

(2) Increasing levels of native lowland hay in the ration decreased total gas and total acid production in rumen samples.

(3) Pelleting of the rations had no effect on the amount of gas or acid produced in manometric fermentations of samples of rumen contents.

III. Effects of Pelleting and Level of Lowland Hay on Relative Production and Proportions of Volatile Fatty Acids in the Rumen

Status of the Problem

A large part of the energy requirements of ruminants is supplied by VFA produced by fermentation of ingested feed in the rumen. Thus, the value of a feed as a source of energy for ruminants depends, to a considerable extent, upon the amount of VFA produced and the efficiency with which they are utilized.

Efficiency of utilization of VFA as a source of energy by lambs is related to the relative proportions of individual VFA produced. Acetic acid is normally the predominant fatty acid produced in the rumen, with lesser amounts of propionic, butyric and other VFA. When the proportion of acetic acid to other VFA is decreased, more efficient utilization of gross energy occurs.

Since it has been noted that the proportions of individual VFA produced may be influenced by ration variations and ration treatment, a study was undertaken to determine the effects of pelleting rations

containing varying levels of native lowland hay on the levels of VFA in rumen contents.

Trial 1

Object

To determine the effects of pelleting rations containing varying levels of native lowland hay on relative production and proportions of VFA in rumen contents of fattening lambs.

Experimental

Samples of rumen contents were obtained from four lambs from each lot (3 lambs in Lot 6) in the second feeding trial, using the same technique as described previously (p. 39). Portions of each homogenized and strained rumen sample were transferred to test-tubes. Fermentation was stopped by addition of 50 per cent H_2SO_4 (v/v) equal to three per cent of the sample volume (Emery *et al*., 1956). The tubes were tightly stoppered and stored at -5° C. until analyzed.

Acetic, propionic, butyric, and valeric and higher acid fractions in rumen liquid were separated and estimated by direct partition chromatography by the method of Keeney (1955), as outlined by Asplund (1957).

Results and Discussion

Average concentrations of VFA found in the rumen samples are summarized in Table 12 and illustrated graphically in Fig. 4. Analyses of variance of the data and significant mean differences are shown in Appendix D, Tables I and II.

Molar concentrations of VFA in rumen liquid were influenced to some extent by the inclusion of lowland hay in the rations. Among lambs fed non-pelleted rations, lower levels of total VFA were obtained for those fed lowland hay rations (Lots 2, 4 and 6) than for those fed alfalfa and

barley (Lot 1), but the difference was significant only for Lot 1 vs. Lot 6 (alfalfa-barley vs.90 per cent lowland hay).

Table 12. Effects of Pelleting and Level of Lowland Hay on Molar Concentrations of Volatile Fatty Acids: Trial 1
(mM./100 ml. rumen liquid)

Hours After Feeding Lambs	Alfalfa-Barley	45% Lowland Hay		60% Lowland Hay		90% Lowland Hay	
		Chopped	Pelleted	Chopped	Pelleted	Chopped	Pelleted
Lot No.	1	2	3	4	5	6	7
Total VFA							
0	6.95	4.88	4.37	6.10	6.00	5.17	5.18
1½	9.61	6.63	11.24	6.80	8.22	6.91	5.83
3	7.73	7.51	9.65	7.72	9.17	7.05	7.07
6	7.25	6.85	8.48	6.29	7.19	5.93	5.54
Av.	7.89	6.47	8.44	6.73	7.65	6.26	5.91
Acetic Acid							
0	3.80	3.21	2.72	4.04	3.97	3.75	3.75
1½	5.09	4.33	7.24	4.48	5.32	4.97	3.98
3	4.52	4.60	6.05	5.05	5.98	5.15	5.14
6	4.45	4.49	5.28	4.15	4.84	4.29	3.87
Av.	4.46	4.16	5.32	4.43	5.03	4.54	4.19
Propionic Acid							
0	2.00	0.86	0.76	0.94	1.00	0.94	0.89
1½	2.54	1.37	1.91	1.35	1.52	1.28	1.17
3	1.82	1.81	1.91	1.38	1.60	1.28	1.26
6	1.58	1.29	1.63	1.16	1.20	1.07	0.98
Av.	1.99	1.33	1.55	1.21	1.33	1.14	1.07
Butyric Acid							
0	0.88	0.60	0.67	0.65	0.82	0.35	0.42
1½	1.67	0.75	1.68	0.80	1.08	0.52	0.55
3	1.07	0.92	1.44	0.98	1.35	0.52	0.57
6	0.93	0.88	1.34	0.78	0.98	0.45	0.49
Av.	1.14	0.79	1.28	0.80	1.06	0.46	0.51
Valeric and Higher Acids							
0	0.27	0.21	0.22	0.48	0.21	0.13	0.12
1½	0.30	0.18	0.42	0.18	0.30	0.15	0.14
3	0.33	0.18	0.25	0.30	0.23	0.10	0.10
6	0.29	0.19	0.24	0.20	0.17	0.12	0.20
Av.	0.30	0.19	0.28	0.29	0.23	0.12	0.14

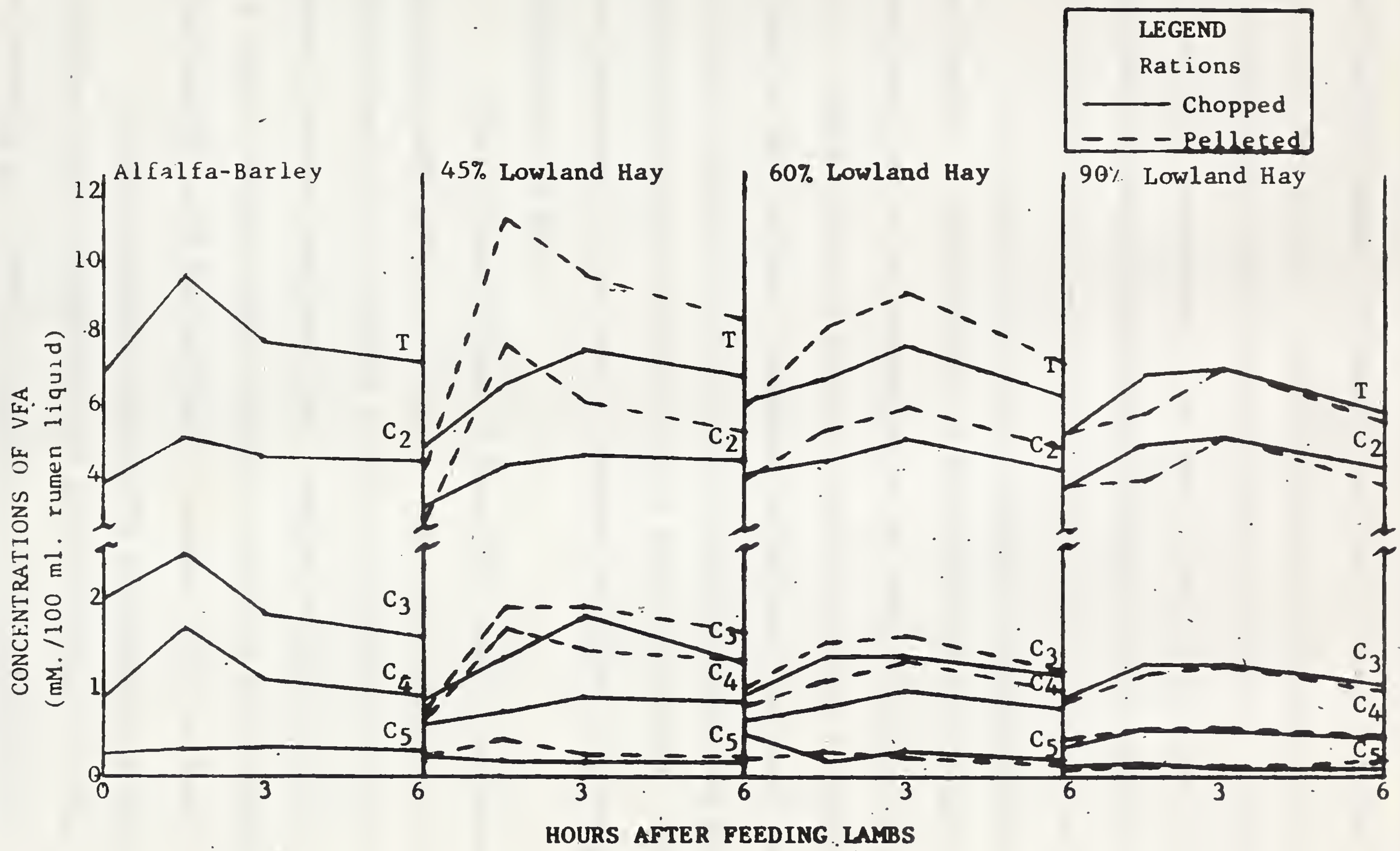


Figure 4. Effects of Pelleting and Level of Lowland Hay on Molar Concentrations of Volatile Fatty Acids in Rumen Fluid: Trial 1
(T=Total VFA ; C_2=Acetate; C_3=Propionate; C_4=Butyrate; C_5=Valeric and Higher acids)

Levels of acetic acid were not affected by lowland hay rations (Lots 2, 4 and 6), but concentrations of propionic and butyric acids were significantly reduced, as compared to those in rumen samples from lambs fed alfalfa and barley. Increasing levels of lowland hay in the ration did not appreciably affect concentrations of acetic or propionic acids, but the ration containing 90 per cent of lowland hay (Lot 6) resulted in significantly lower concentrations of butyric and of valeric and higher acids than did rations with 45 and 60 per cent of lowland hay (Lots 2 and 4).

Pelleting of the rations affected the concentrations of VFA in rumen samples. Concentrations of total VFA were significantly increased by pelleting the ration containing 45 per cent of lowland hay (Lot 3 vs. Lot 2). Higher levels of acetic, propionic and butyric acids accounted for most of the increase. A similar effect was noted with the 60 per cent lowland hay rations (Lot 5 vs. Lot 4), but the differences obtained, as a result of pelleting, were not as large (non-significant). Pelleting the ration containing 90 per cent of lowland hay had no appreciable effect on concentrations of VFA.

Molar concentrations of total VFA, acetic, propionic and butyric acids were significantly affected by the time of sampling of rumen contents. Samples taken prior to feeding (0 hour) contained low levels of acetic, propionic, butyric and total VFA. Samples obtained following feeding (1½ and 3 hours) showed a marked increase in each of the components. Levels of all but butyric acid had declined in samples taken six hours after feeding.

If molar concentrations of VFA in rumen liquid are an indication of relative rates of production and absorption from the rumen, as suggested by McCarthy _et al_. (1958), it appears that the alfalfa-barley ration (Lot 1), and non-pelleted rations containing 45 and 60 per cent of lowland hay, supplied similar amounts of energy as VFA. Lower amounts of energy

were supplied as VFA by rations containing 90 per cent of lowland hay. It is also apparent that pelleting of the 45 per cent lowland hay ration increased the supply of energy from VFA, but pelleting of rations containing 60 and 90 per cent of lowland hay did not appreciably affect the amount of energy supplied from this source. Rate of gain and efficiency of utilization of energy noted in the feeding trial (Table 5) did not correspond with the concentrations of VFA in rumen samples observed above.

The effects of treatment on molar proportions (per cent of total VFA) of VFA in rumen samples are presented in Table 13 and represented graphically in Fig. 5. Analyses of variance of the data and significant mean differences are shown in Appendix D, Tables I and III.

The inclusion of lowland hay in the ration had marked effects on the proportions of VFA in rumen liquid. Acetic acid was increased from 58 per cent on the alfalfa-barley ration (Lot 1), to 64 and 66 per cent of the total VFA on non-pelleted rations containing 45 and 60 per cent of lowland hay respectively (Lots 2 and 4). A significant decrease in the proportion of propionic acid accounted for most of the increase in the proportion of acetic acid; propionic acid decreased from 25 per cent (Lot 1) to 20 and 18 per cent of the total VFA with the respective levels of lowland hay. The non-pelleted ration containing 90 per cent of lowland hay resulted in a further significant increase in the percentage of acetic acid to 72 per cent of the total VFA. However, this increase in acetic acid was not associated with a further decrease in the percentage of propionic acid, but with a five per cent reduction in the percentage of butyric acid (significant).

Pelleting of the rations did not have marked effects on molar proportions of VFA in rumen liquid. The percentages of acetic and propionic acids decreased slightly, and butyric acid increased slightly when

the rations were pelleted. However, differences were small; the only significant change was an increase in the percentage of butyric acid as a result of pelleting the 45 per cent lowland hay ration. Consequently, it may be concluded that pelleting had no effect on efficiency of utilization of the energy supplied as VFA from the rumen.

Table 13. Effects of Pelleting and Level of Lowland Hay on Molar Proportions of Volatile Fatty Acids: Trial 1
(Per cent of Total VFA)

Hours After Feeding Lambs	Alfalfa -Barley	45% Lowland Hay		60% Lowland Hay		90% Lowland Hay	
		Chopped	Pelleted	Chopped	Pelleted	Chopped	Pelleted
Lot No.	1	2	3	4	5	6	7
Acetic Acid							
0	54.8	66.0	61.8	66.5	66.0	72.3	72.5
1½	53.2	65.4	64.4	65.9	64.7	71.8	67.2
3	58.9	61.2	63.0	65.6	65.3	73.1	72.8
6	64.0	65.0	61.3	66.2	67.4	72.0	69.6
Av.	57.7	64.4	62.6	66.1	65.8	72.3	70.5
Propionic Acid							
0	29.1	17.3	18.4	15.4	16.7	18.4	16.3
1½	26.5	20.6	17.4	19.9	18.4	18.4	19.5
3	23.3	23.6	19.9	17.8	17.6	18.0	17.9
6	20.6	19.0	19.6	18.4	16.6	18.2	17.9
Av.	24.9	20.1	18.8	17.9	17.3	18.3	17.9
Butyric Acid							
0	12.4	11.6	15.0	11.2	13.6	6.8	8.0
1½	17.1	11.3	14.6	11.6	13.2	7.6	10.0
3	13.6	12.7	14.4	12.6	14.6	7.3	7.9
6	11.6	13.1	16.0	12.2	13.8	7.6	9.0
Av.	13.7	12.2	15.0	11.9	13.8	7.3	8.7
Valeric and Higher Acids							
0	3.7	5.0	4.8	7.0	3.8	2.5	3.2
1½	3.2	2.7	3.7	2.6	3.6	2.2	3.3
3	4.1	2.6	2.7	4.0	2.6	1.5	1.4
6	3.8	2.9	3.1	3.1	2.3	2.1	3.6
Av.	3.7	3.3	3.6	4.2	3.0	2.1	2.8

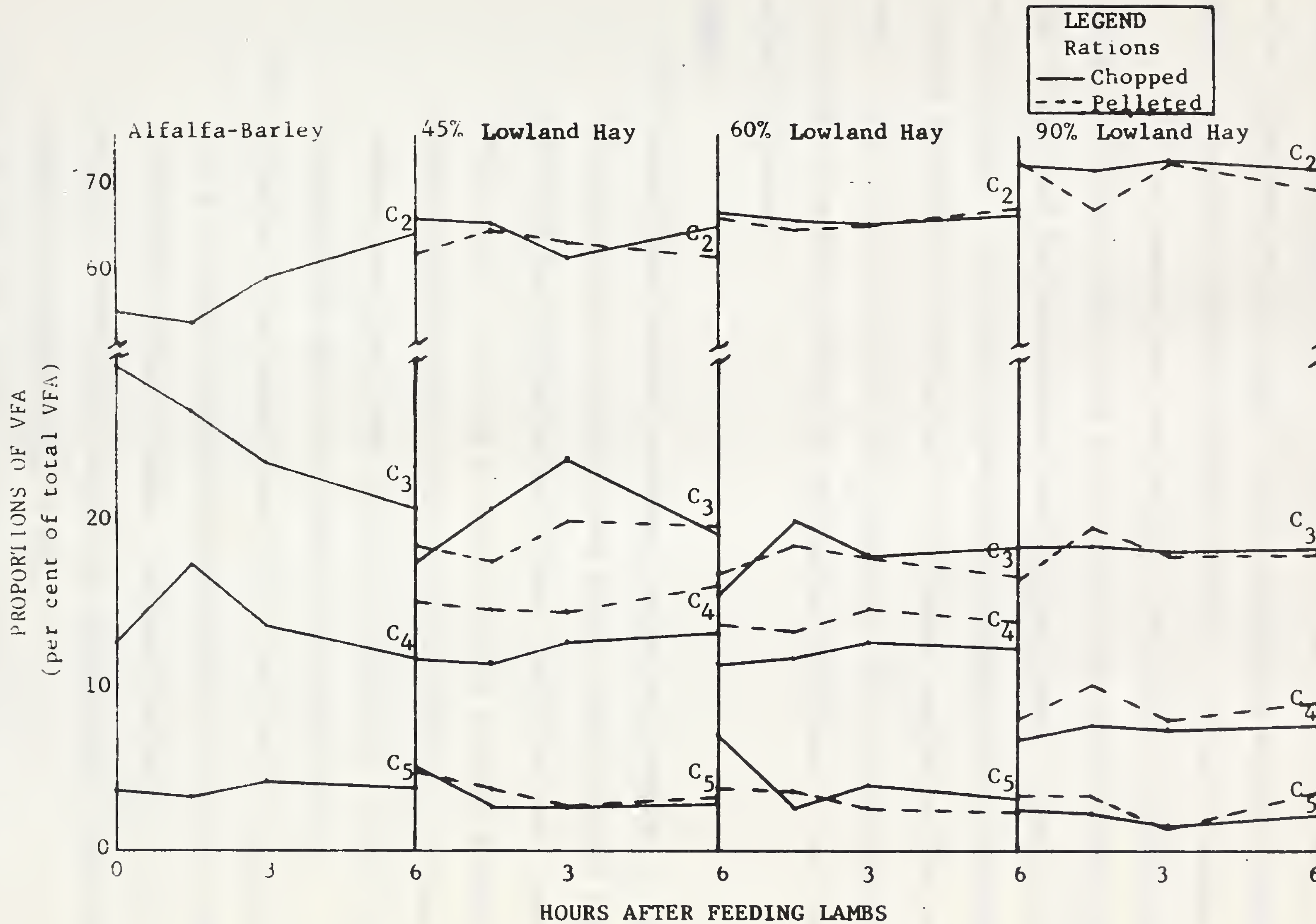


Figure 5. Effects of Pelleting and Level of Lowland Hay on Molar Proportions of Volatile Fatty Acids in Rumen Fluid: Trial 1
(C_2=Acetate; C_3=Propionate; C_4=Butyrate; C_5=Valeric and Higher Acids.)

The proportions of VFA were not affected by time of sampling. There was little change in the molar proportions of the individual VFA found in rumen samples taken at 0, 1½, 3 or 6 hours after feeding. This agrees with other studies (Lewis, 1961), in which it was noted that the molar proportions of the rumen VFA remained relatively constant throughout the day.

It is apparent that the levels of VFA found in samples of rumen contents of lambs in this trial do not reflect the marked differences in rate of gain and feed consumption noted between groups of lambs fed the various rations. It is also evident that pelleting of the rations did not increase efficiency of utilization of the energy supplied as VFA from the rumen. These results indicate that analyses of VFA in rumen contents do not provide an accurate method for predicting differences in the productive value of a ration.

Summary

(1) Non-pelleted rations containing lowland hay resulted in lower concentrations of propionic and butyric acids in rumen liquid than did the alfalfa-barley ration. The 90 per cent lowland hay ration (Lot 6) resulted in less total VFA in rumen samples than did the control ration (Lot 1), and less butyric acid than the rations containing lower levels of lowland hay. Acetic acid concentrations were not affected by the rations used.

(2) Pelleting of the ration containing 45 per cent of lowland hay (Lot 3 vs. Lot 2) increased concentrations of total VFA because of increased levels of acetic, propionic and butyric acids. Pelleting of the other lowland hay rations had no significant effects on concentrations of VFA.

(3) Rations containing 45 and 60 per cent of lowland hay resulted in higher proportions of acetic acid and lower proportions of propionic acid than did the alfalfa-barley ration. As the level

of lowland hay in the ration was increased to 90 per cent, the proportion of acetic acid continued to increase and the proportion of butyric acid decreased.

(4) Pelleting of the rations had no appreciable effects on the proportions of VFA in rumen liquid.

(5) The concentrations and proportions of VFA in rumen samples did not reflect differences in rate of gain or feed consumption of lambs in the trial (Table 5). Consequently, analyses of VFA in rumen contents do not appear to be an accurate method for predicting the relative value of rations for productive purposes.

Trial 2

Object

Since rumen samples obtained by stomach tube may be diluted with saliva, a trial was designed to determine concentrations of VFA in rumen samples obtained via fistulae from wethers fed pelleted and non-pelleted rations containing varying levels of native lowland hay.

Experimental

Rumen samples were obtained from the five wethers in the fourth feeding trial, in the same manner as described previously (p. 45). Samples from each wether were taken three hours after feeding, on the 14th and 21st days of the pretreatment period, and on the 1st, 7th, 14th, 21st and 28th days of each of the two replicates of the trial.

The samples were strained, acidified and analyzed immediately for VFA by the methods described in Trial 1 of this experiment.

A Photovolt Portable Electronic pH Meter was used to determine pH of rumen samples as soon as they were collected.

Results and Discussion

Molar concentrations of VFA in rumen samples are summarized in Table 14 and depicted graphically in Fig. 6. Analyses of variance of the data and significant mean differences are presented in Appendix D, Tables IV and V.

The wethers appeared to have adjusted to the alfalfa-barley ration by the time the first samples were taken in the pretreatment period. Average molar concentrations of VFA in rumen liquid decreased between 14 and 21 days, but the differences were small (non-significant). There was very little variation between wethers in levels of VFA in rumen samples.

The rumen microflora apparently required a period of time in which to adjust to changes in the rations fed to the wethers. There was a marked decrease in molar concentrations of VFA in rumen liquid on the first day after the wethers were fed the lowland hay rations, as compared to those in rumen liquid from wethers fed the alfalfa-barley ration, but substitution of pelleted for non-pelleted lowland hay rations (or the reverse) had only slight effects on concentrations of VFA. This indicates that a greater adjustment occurred in the microbial population in the rumen when lowland hay rations were fed in place of the alfalfa-barley ration, than when the physical form of the lowland hay rations was changed. Adjustment of the microflora, as measured by concentrations of VFA in the rumen, appeared to be complete within 14 days after commencing to feed the various rations. No significant differences were found in concentrations of VFA in rumen samples taken on the 14th, 21st, and 28th days of the trial.

In general, the effects of treatments on concentrations of VFA were similar to those obtained in Trial 1, but some differences were noted. Lower levels of VFA were found in rumen samples from sheep fed non-pelleted lowland hay rations (Lots 4 and 6), than in those from sheep fed the

Table 14. Effects of Pelleting and Level of Lowland Hay on pH of Rumen Contents and Molar Concentrations of Volatile Fatty Acids: Trial 2

Days on Test Ration	Pre-treatment	Alfalfa-Barley	60% Lowland Hay Chopped	60% Lowland Hay Pelleted	90% Lowland Hay Chopped	90% Lowland Hay Pelleted
Lot No.		1	4	5	6	7
Rumen pH		5.8	6.5	5.8	6.7	6.2
Concentrations of VFA (mM./100 ml. rumen liquid)						
Total VFA						
1		13.85	7.79	7.36	7.08	4.92
7		11.80	9.75	10.40	6.68	7.16
14	15.09	12.68	9.06	15.52	8.40	9.64
21	13.34	15.79	8.92	14.02	8.44	11.94
28		12.44	10.40	12.00	8.22	12.32
Av.	14.22	13.31	9.18	11.86	7.76	9.19
Acetic Acid						
1		7.76	4.46	4.72	4.26	3.16
7		6.92	6.30	6.20	4.48	4.40
14	8.50	7.14	5.57	9.48	5.47	6.14
21	7.49	8.98	5.65	8.78	5.42	7.54
28		6.94	6.55	7.40	5.34	7.92
Av.	8.00	7.54	5.71	7.31	5.00	5.83
Propionic Acid						
1		3.24	1.88	1.44	1.47	0.92
7		2.61	1.74	2.52	1.24	1.79
14	3.06	2.66	1.90	2.70	1.70	2.36
21	2.97	3.46	1.82	3.05	1.90	2.82
28		2.96	2.36	2.56	1.86	2.46
Av.	3.01	2.99	1.94	2.46	1.63	2.07
Butyric Acid						
1		2.31	1.12	0.94	0.90	0.61
7		1.84	1.46	1.50	0.76	0.76
14	2.55	2.39	1.30	2.88	1.00	0.91
21	2.15	2.62	1.20	1.68	0.87	1.30
28		2.12	1.24	1.54	0.80	1.64
Av.	2.35	2.25	1.27	1.71	0.87	1.04
Valeric and Higher Acids						
1		0.54	0.33	0.26	0.44	0.24
7		0.44	0.24	0.18	0.21	0.22
14	0.98	0.50	0.28	0.46	0.23	0.23
21	0.73	0.78	0.24	0.52	0.24	0.28
28		0.42	0.24	0.50	0.22	0.30
Av.	0.86	0.53	0.27	0.38	0.27	0.25

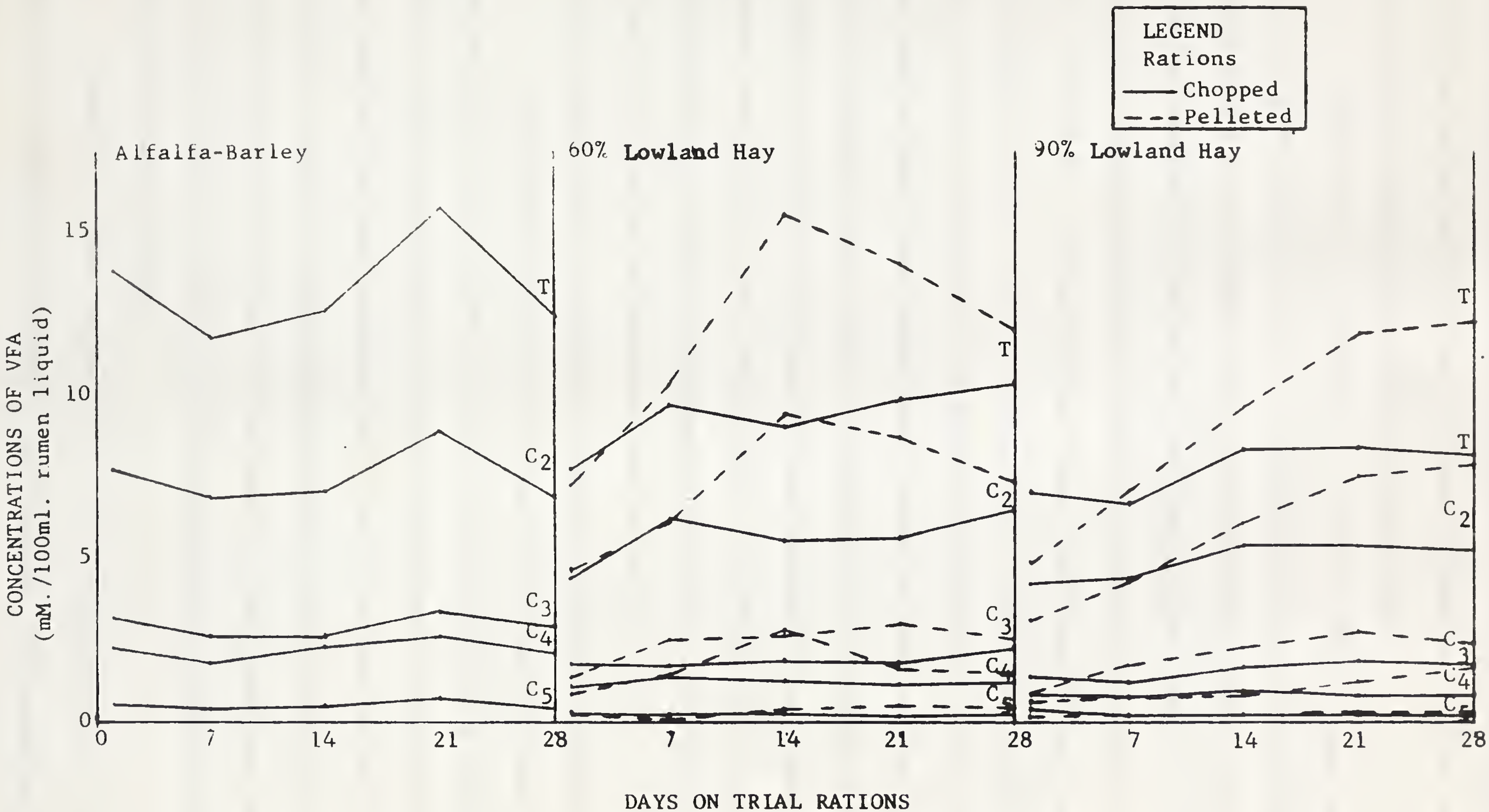


Figure 6. Effects of Pelleting and Level of Lowland Hay on Molar Concentrations of Volatile Fatty Acids in Rumen Fluid: Trial 2

(T=Total VFA; C_2=Acetate; C_3=Propionate; C_4=Butyrate; C_5=Valeric and Higher Acids)

alfalfa-barley ration (Lot 1); the ration containing 90 per cent of lowland hay resulted in the lowest concentration of VFA. The level of acetic acid was not affected by the non-pelleted lowland hay rations in Trial 1, whereas it was significantly reduced in this trial.

The effects of pelleting on levels of VFA in rumen samples were similar to that noted in Trial 1. Pelleting of the rations increased molar concentrations of total VFA, acetic, propionic and butyric acids in rumen liquid, as compared to similar non-pelleted rations, but the increases were significant only when the 60 per cent lowland hay ration was pelleted (Lot 5 vs. Lot 4). In the previous trial, pelleting did not have significant effects at either level of lowland hay. However, it should be noted that in Trial 1 (Table 12), the difference in levels of total VFA, in rumen samples taken three hours after feeding of pelleted and non-pelleted rations containing 60 per cent of lowland hay, was as great as that observed in this trial, and the difference occurred because of increased acetic, propionic and butyric acids. Pelleting of the 90 per cent lowland hay ration had greater effects on concentrations of VFA in this trial, but in neither trial were the differences found to be significant. Consequently, it would appear that the results of both trials were very similar.

The failure of two of the wethers to consume pelleted rations during the first week of the trial may have influenced the concentrations of VFA. Lower concentrations of total VFA and acetic acid were found in rumen samples from these wethers (Lots 5 and 7) on the 1st and 7th days of Replicate 1 than in corresponding samples from wethers fed the same rations in Replicate 2. This was indicated by the significant effects obtained for replicates and for the interactions of replicates with treatments and with days on test.

The pH of rumen contents (Table 14) varied slightly with different rations, and appeared to reflect, to some extent, the concentrations of VFA.

Lowland hay in non-pelleted rations at levels of 60 and 90 per cent (Lots 4 and 6), resulted in higher pH than did the control ration (Lot 1), reflecting the lower concentrations of VFA on the lowland hay rations. Pelleting of the rations resulted in increased VFA levels and lower pH in the rumen contents.

Average molar proportions of VFA (per cent of total VFA) in rumen liquid are shown in Table 15 and represented graphically in Fig. 7. Analyses of variance of the data and significant mean differences are summarized in Appendix D, Tables VI and VII.

The proportions of VFA in rumen samples were significantly affected by the addition of lowland hay to the ration. A lower percentage of acetic acid and a higher percentage of butyric acid were obtained on the alfalfa-barley ration (Lot 1), than on rations containing lowland hay. The proportions of propionic and of valeric and higher acids were not affected by the different rations. Increasing the level of lowland hay from 60 to 90 per cent of the ration resulted in a slight increase in the percentage of acetic acid, and a slight decrease in the percentage of butyric acid, but the differences were not significant.

Pelleting of the rations had no appreciable effect on the proportions of VFA in rumen liquid. This is in complete agreement with the results obtained in the previous trial, and indicates that pelleting had no effect on efficiency of utilization of the energy supplied as VFA.

The results noted above do not agree entirely with those obtained in the previous trial, although the results of both trials indicate similar effects on energy utilization. It was observed in Trial 1 that the proportion of propionic acid was affected by rations containing 60 per cent of lowland hay, and the proportion of butyric acid was affected only by rations containing 90 per cent of lowland hay; this is different from the results noted above. It is also apparent that the 90 per cent lowland hay rations

resulted in a greater increase in the percentage of acetic acid, and a greater decrease in the percentages of propionic and butyric acids in Trial 1 than in this trial. Nevertheless, the results of both trials indicate that the addition of lowland hay to the rations increased the proportions of acetic acid at the expense of propionic and/or butyric acids, suggesting less efficient utilization of the energy supplied as VFA than with the alfalfa-barley ration.

Table 15. Effects of Pelleting and Level of Lowland Hay on Molar Proportions of Volatile Fatty Acids: Trial 2
(Per cent of Total VFA)

Days on Test Ration	Pre-treatment	Alfalfa -Barley	60% Lowland Hay		90% Lowland Hay	
			Chopped	Pelleted	Chopped	Pelleted
Lot No.		1	4	5	6	7
Acetic Acid						
1		56.0	57.3	62.6	61.3	60.7
7		58.6	64.8	59.6	67.0	62.0
14	56.4	56.4	61.4	61.0	65.0	64.3
21	56.6	56.6	63.3	62.4	64.3	63.2
28		55.8	63.0	60.8	65.1	64.3
Av.	56.5	56.7	62.0	61.3	64.5	62.9
Propionic Acid						
1		23.2	25.2	19.5	20.4	17.4
7		22.2	17.9	25.9	18.4	22.3
14	20.1	20.8	21.0	17.4	20.2	23.8
21	22.1	22.0	20.4	22.0	22.4	23.6
28		24.0	22.8	23.0	22.4	20.0
Av.	21.1	22.4	21.5	21.6	20.8	21.4
Butyric Acid						
1		16.9	13.3	11.8	12.8	15.1
7		15.5	14.9	13.0	11.4	12.6
14	16.9	18.9	14.4	18.7	12.0	9.6
21	15.8	16.4	13.6	12.0	10.4	10.9
28		16.8	12.0	12.4	9.8	13.2
Av.	16.4	16.9	13.6	13.6	11.2	12.3
Valeric and Higher Acids						
1		4.0	4.2	6.2	5.4	6.8
7		3.8	2.4	1.4	3.2	3.2
14	6.6	3.8	3.2	3.0	2.9	2.4
21	5.5	5.0	2.8	3.6	2.9	2.2
28		3.3	2.4	3.8	2.8	2.4
Av.	6.0	4.0	3.0	3.6	3.4	3.4

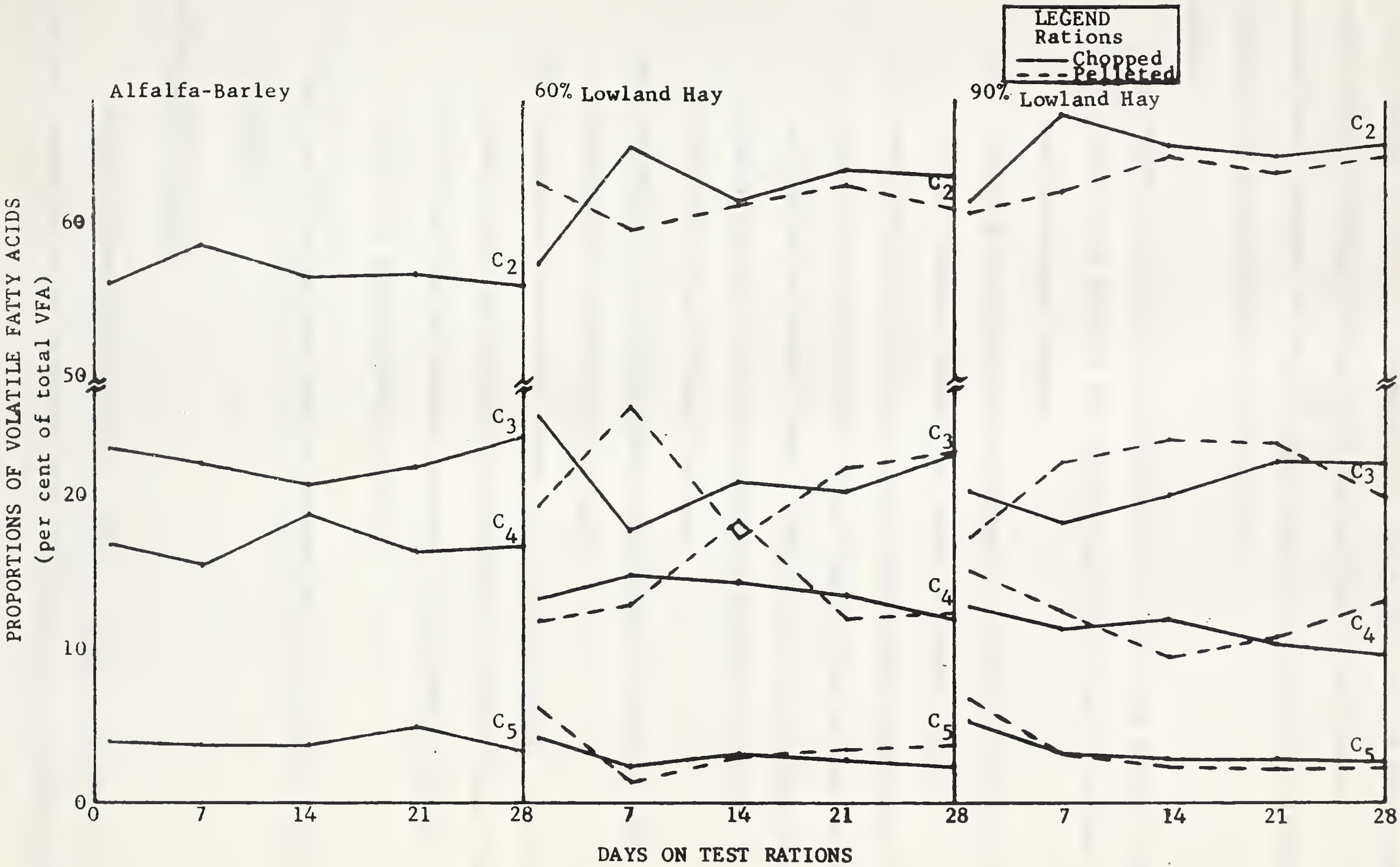
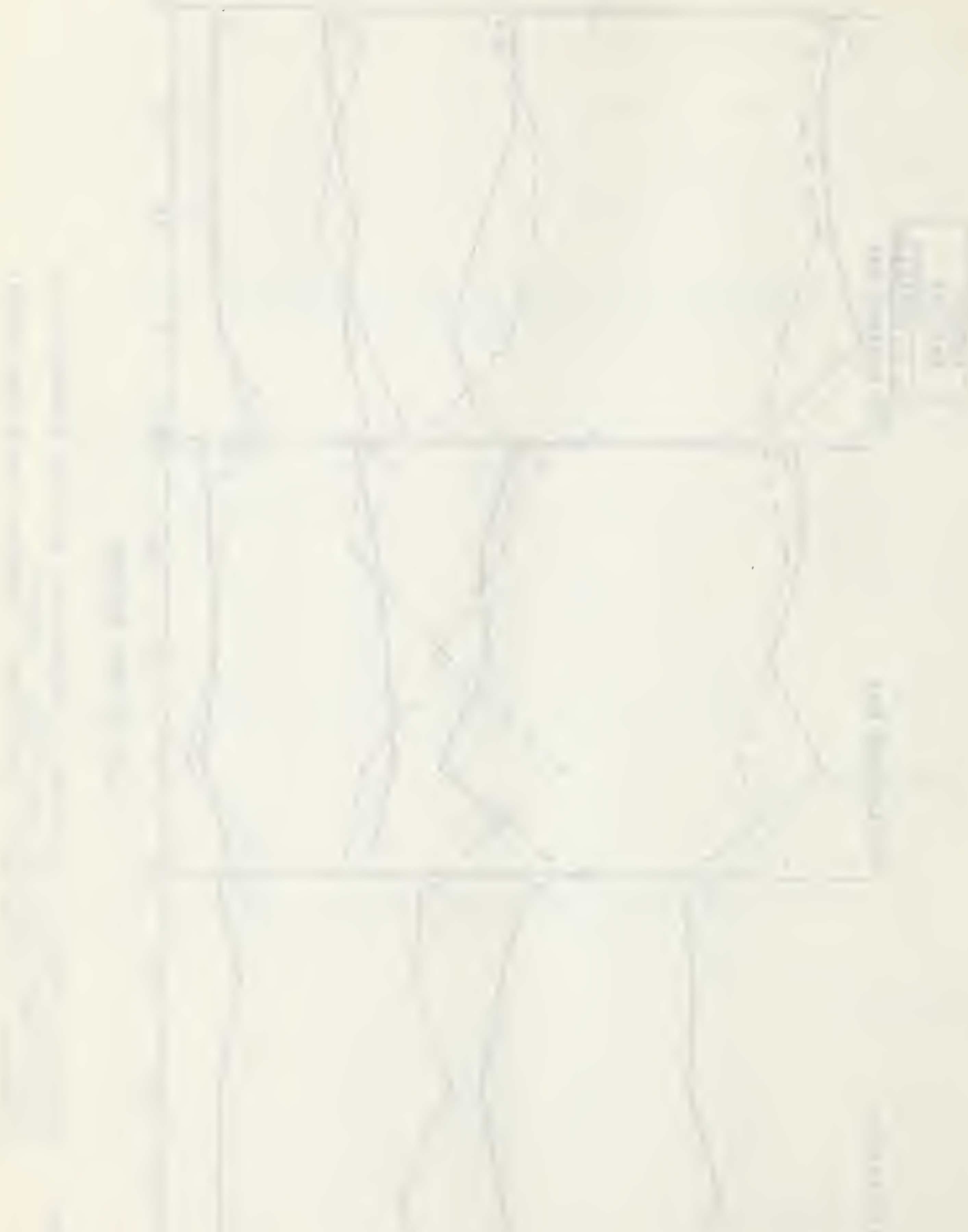


Figure 7. Effects of Pelleting and Level of Lowland Hay on Molar Proportions of Volatile Fatty Acids in Rumen Fluid: Trial 2
(C_2=Acetate; C_3=Propionate; C_4=Butyrate; C_5=Valeric and Higher Acids)



Since the results of the two trials agree fairly closely, indicating similar effects on concentrations and proportions of VFA in rumen samples from pelleting and levels of lowland hay in the rations, it seems apparent that analyses of VFA in rumen samples do not reflect differences that exist in the productive value of rations.

Summary

(1) Significantly higher concentrations of VFA were found in rumen liquid from wethers fed alfalfa and barley than from those fed the lowland hay rations.

(2) Pelleting of the rations increased concentrations of total VFA, acetic, propionic and butyric acids in rumen liquid.

(3) Lower proportions of acetic acid, and higher proportions of butyric acid were found in rumen samples from wethers fed the alfalfa-barley ration, than in those from wethers fed the lowland hay rations. The percentage of propionic acid was not affected by the different rations.

(4) Pelleting of the rations had no appreciable effects on the proportions of VFA in rumen liquid.

(5) In general, the results agreed with those obtained in the previous trial, and indicated that analyses of VFA in rumen samples do not provide an accurate measure of differences between rations in their productive value.

IV. Effects of Pelleting and Level of Lowland Hay on Cellulose Digestion in the Rumen

Status of the Problem

Ground roughage and decreased amounts of long roughage in the ration are associated with increased rate of passage of food through the

digestive tract, and decreased digestibility, particularly of the fibrous part of the ration. Consequently, an experiment was designed to determine the effects of grinding and pelleting of high-roughage rations on cellulose digestion in the rumen.

Object

To determine the effects of pelleting and level of lowland hay on cellulose digestion in the rumen.

Experimental

The 'nylon bag' technique (el-Shazly *et al*., 1961) was used to determine cellulose digestion in the rumen. Bags (2½" x 5") were made from terylene cloth (150 x 100 threads/sq. in.). One gram of shredded filter paper was placed in each bag and six of these tied to a length of nylon fish-line.

During the last week of each replicate of the fourth feeding trial, previously outlined, a string of six bags was placed in the rumen of each wether via the fistula. The wethers had been fed 12 hours previously, and were not fed again until removal of all the bags. Three were removed from each rumen after 12 hours, and three after 24 hours, rinsed in water to remove adhering rumen ingesta, and the contents washed into 90 ml. centrifuge tubes.

Cellulose was determined by the method outlined by Crampton and Maynard (1938) and modified by Donefer *et al*. (1960).

Results and Discussion

Data obtained on percentage cellulose digested in the rumen are shown in Table 16. It may be noted that an incubation period of 12 hours resulted in a much smaller percentage digestion of the cellulose in all lots, than in a 24 hour incubation period. This could partially be caused by time required for the bags to mix with the rumen contents and become

saturated with rumen liquid.

Table 16. Effects of Pelleting and Level of Lowland Hay on Percentage Cellulose Digestion in the Rumen

Hours of Incubation	Alfalfa -Barley	60% Lowland Hay		90% Lowland Hay	
		Chopped	Pelleted	Chopped	Pelleted
Lot No.	1	4	5	6	7
12	6.85	10.83	5.13	7.78	4.70
24	29.68	54.54	28.07	44.66	57.77

The non-pelleted rations containing lowland hay (Lots 4 and 6) resulted in a much higher percentage of cellulose digestion, particularly after 24 hours of incubation, than did the alfalfa-barley ration (Lot 1). This indicates that the higher levels of roughage in the lowland hay rations resulted in a greater cellulolytic activity of the rumen micro-organisms, as compared to the alfalfa-barley ration. However, the 90 per cent lowland hay ration (Lot 6) did not result in as high a percentage of cellulose digestion as the 60 per cent lowland hay ration (Lot 4), indicating a lower cellulolytic activity.

Pelleting of the 60 per cent lowland hay ration decreased cellulose digestion in incubation periods of 12 and 24 hours. Pelleting of the 90 per cent lowland hay ration had the same effect in a 12 hour incubation period, but after 24 hours of incubation, cellulose digestion was greater than that obtained with the same non-pelleted ration. The increased cellulose digestion, as a result of pelleting this ration, is just the reverse of the results obtained with the 60 per cent lowland hay ration, and cannot be explained.

Although this technique has been used with some success in rumen studies (el-Shazly _et al_., 1961; Belasco _et al_., 1958; Erwin and Elliston,

1959), a standard procedure has not yet been developed. Archibald *et al.* (1961) were not able to determine differences in digestion with this method.

Summary

(1) Non-pelleted lowland hay rations resulted in greater cellulose digestion in the rumen than did the alfalfa-barley ration. Increasing the level of lowland hay from 60 per cent to 90 per cent of the ration reduced cellulose digestion.

(2) Pelleting of the 60 per cent lowland hay ration reduced cellulose digestion, whereas pelleting of the 90 per cent lowland hay ration increased cellulose digestion, as compared to similar non-pelleted rations.

GENERAL DISCUSSION

It was evident that pelleting greatly increased the nutritive value of rations containing lowland hay. Rate of gain, daily feed consumption and efficiency of feed utilization by lambs were markedly improved by pelleting of the rations. These results were similar to those of Wallace and Hubbert (1959), who found increased rate of gain and feed intake of steers fed pelleted rations of native meadow hay.

The low quality of the lowland hay was indicated by very low digestion coefficients obtained for dry matter, crude protein and gross energy in rations containing high levels of lowland hay. Increased digestibility was noted in rations containing lower levels of lowland hay, but it did not equal that of rations of alfalfa and barley. Pelleting of the lowland hay rations increased digestibility of dry matter and crude protein in rations containing 90 per cent of lowland hay, but did not improve, and even decreased digestibility in rations containing lower levels of lowland hay. It appears that increased feed intake was largely responsible for the improved performance of fattening lambs fed pelleted rations.

Pelleting of the lowland hay rations eliminated interference of wild barley (*Hordeum jubatum*). Non-pelleted rations containing wild barley were consumed reluctantly by lambs, but the pelleted rations were readily consumed and feed wastage was eliminated. Similar effects were noted by Wallace *et al*. (1961). It was apparent that increased feed intake, as a result of pelleting, markedly improved the value of low-quality hay for lambs. Native lowland hay, at levels as high as 84 per cent of the ration, was fed to fattening lambs with excellent results when the rations were pelleted. Similar improvements in feeding value by pelleting of low-quality roughages have been noted in other studies (see Literature Review).

Rate of fermentation in rumen samples was not affected by pelleting of the rations. This does not agree with the results obtained by Meyer et al. (1959a), who noted increased rate of fermentation when high-quality alfalfa hay was pelleted. However, in the results reported here, similar rates of fermentation were found on pelleted and non-pelleted rations containing the same level of lowland hay. Decreased rate of fermentation, with increasing levels of lowland hay in the ration, did not reflect small but significant differences in digestibility or quality of the rations, and was not associated with the differences noted in feed intake. Consequently, a measurement of the rate of fermentation in rumen samples could not be used to indicate differences between rations in productive value.

The concentrations of VFA in rumen samples did not provide an accurate measure of the value of a ration. Levels of VFA in rumen liquid were increased by pelleting of the 45 per cent lowland hay ration, and possibly by pelleting of the 60 per cent lowland hay ration. Pelleting of the 90 per cent lowland hay rations had no significant effects on concentrations of VFA. The results indicate that concentrations of VFA in rumen fluid did not reflect the differences found in rate of gain and feed intake of sheep fed the pelleted and non-pelleted rations.

It was noted that increasing levels of lowland hay in the ration increased the proportions of acetic acid and decreased the proportions of propionic and/or butyric acids in rumen liquid. Consequently, efficiency of utilization of energy supplied as VFA should decrease with increasing levels of roughage in the ration (see Literature Review). Pelleting of the rations had no appreciable effects on the proportions of the individual VFA, and should not have affected efficiency of utilization of energy. This agrees with the results obtained by Leffel (1960), who noted that

pelleting of timothy hay had no appreciable effects on levels or proportions of VFA.

It has been suggested that analyses of VFA in rumen fluid might be useful in appraising the value of a ration. Shaw et al. (1959) observed that "analysis of rumen fluid for VFA may be a valuable guide for evaluation of dietary regimens for their probable effect on the fat content of milk." Shaw (1959) suggested the technique could be used to select rations which might be expected to be most efficient for fattening. However, the results reported here demonstrate that analyses of VFA in rumen fluid do not give an accurate measurement of the value of a ration for productive purposes.

Since increases in rate of fermentation and levels or proportions of VFA in the rumen contents were not of sufficient magnitude to account for the increased gain of lambs noted when rations were pelleted, one is forced to the conclusion that they gained faster because they consumed more feed.

GENERAL SUMMARY

Four experiments were conducted to study the effects in sheep of pelleting of rations containing varying levels of native lowland hay on rate of gain and efficiency of feed utilization, rate of rumen fermentation, relative production and proportions of volatile fatty acids in the rumen, and rate of cellulose digestion in the rumen. A summary of the results is presented below:

I. Effects of Pelleting and Level of Lowland Hay on Rate of Gain and Efficiency of Feed Utilization

Four trials were undertaken to study the effects of pelleting of lowland hay rations on rate of gain and efficiency of feed utilization of lambs. In all trials a control ration of 1/3 long alfalfa and 2/3 whole barley was fed for comparison with the lowland hay rations. In the first trial, wether lambs were fed pelleted and non-pelleted rations containing 45 and 60 per cent of lowland hay, and 60 per cent of lowland hay plus five per cent of stabilized animal fat. It was noted that rate of gain and efficiency of feed utilization of lambs fed non-pelleted rations were comparable to those of lambs fed alfalfa and barley. Pelleting of the rations increased rate of gain and feed intake, but feed and energy conversion were improved only at the 45 per cent level of lowland hay. Increasing the energy content of the 60 per cent lowland hay rations reduced feed intake, but had no effect on rate of gain or energy required per unit of gain.

The second trial was conducted to study the effects of pelleting of rations containing 45, 60 and 90 per cent of lowland hay on rate of gain and efficiency of feed utilization of lambs, and on apparent digestibility of the ration. It was observed that non-pelleted rations were consumed in low amounts and produced unsatisfactory rates of gain. Pelleting the

rations resulted in marked improvements in rate of gain and efficiency of feed utilization; rate of gain of lambs fed pelleted rations containing 45 and 60 per cent of lowland hay was comparable to the high rate of gain of lambs fed alfalfa and barley. Digestion coefficients for dry matter, crude protein and gross energy were similar in non-pelleted rations containing 45 and 60 per cent of lowland hay, but were lower than those obtained with the alfalfa-barley ration. Very low digestion coefficients were obtained in rations containing 90 per cent of lowland hay. Pelleting resulted in decreased digestibility of dry matter and gross energy in the 45 per cent lowland hay ration, had no effect on digestion coefficients in the 60 per cent lowland hay ration, and increased digestibility of dry matter and crude protein in the 90 per cent lowland hay ration. Rations containing lowland hay resulted in a dark discoloration of the rumen epithelium. A marked decrease in number and size of rumen papillae was noted in lambs fed non-pelleted lowland hay rations, but the effect was not as apparent when the rations were pelleted.

The third trial was designed to study the effects of increased available nitrogen from one per cent of urea and 0.1 per cent of each of lysine and methionine on performance of lambs fed pelleted and non-pelleted rations containing 84 per cent of lowland hay. It was noted that lambs self-fed non-pelleted rations containing lowland hay consumed more feed, but gained less weight than lambs hand-fed alfalfa and barley. Pelleting of the lowland hay rations resulted in marked increases in rate of gain, feed intake and efficiency of feed utilization, as compared to similar non-pelleted rations. Supplementation of the lowland hay rations with urea, and with urea and amino acids, did not improve rate of gain of fattening lambs.

In the fourth trial, a study was conducted on the effects of pelleting on digestibility of rations containing 60 and 90 per cent of

lowland hay when fed to mature wethers. It was observed that digestibility of dry matter, crude protein and gross energy was lower in the lowland hay rations than in the alfalfa-barley ration; the greatest reduction in digestibility occurred in the rations with the higher level of lowland hay. Pelleting of the rations did not appreciably affect the digestion coefficients.

The results of the experiment indicated that pelleting of the rations markedly improved the value of lowland hay, and that levels of lowland hay as high as 84 per cent of pelleted rations could be fed to fattening lambs with excellent results. The increase in value of the low-quality roughage was largely the result of increased feed consumption.

II. Effects of Pelleting and Level of Lowland Hay on Rate of Rumen Fermentation

Two trials were undertaken to study the rate of rumen fermentation, as measured by increase in pressure and total production of gas and acid, in manometric fermentations of rumen samples. In the first trial, rumen contents were sampled at varying intervals by stomach tube. It was observed that a high rate of fermentation was obtained with the ration of alfalfa and barley. Rate of fermentation was reduced a similar amount by rations containing 45 and 60 per cent of lowland hay; very low rates of fermentation were obtained with rations containing 90 per cent of lowland hay. Pelleting of the rations had no appreciable effect on rate of fermentation of rumen samples. A lower rate of fermentation was obtained in rumen samples taken just before feeding, than in samples taken 1½, 3 and 6 hours after feeding, with rations containing 45 and 60 per cent of lowland hay. At all sampling intervals, a high rate of fermentation was noted with the alfalfa-barley ration, and a very low rate of fermentation with 90 per cent lowland hay rations.

The second trial was conducted to study the rate of fermentation in rumen samples obtained via fistulae from mature wethers. All samples were taken three hours after feeding. The results were very similar to those obtained in the previous trial. A high rate of fermentation was observed in rumen samples from wethers fed alfalfa and barley, and increasing levels of lowland hay in the ration resulted in decreased rates of fermentation. Pelleting of the rations had no effect on the rate of fermentation.

The results of the experiment indicated that differences in rate of fermentation could not account for the effects on digestibility noted in the feeding trials, and could not explain the increased feed consumption that occurred when rations were pelleted. Rate of fermentation in rumen samples was not a guide to productive value of a ration.

III. Effects of Pelleting and Level of Lowland Hay on Relative Production and Proportions of Volatile Fatty Acids in the Rumen

Two trials were conducted to study the effects of pelleting of lowland hay rations on concentrations and proportions of VFA in rumen fluid of sheep. In the first trial, rumen contents were sampled by stomach tube at varying intervals after feeding. It was noted that non-pelleted rations containing lowland hay resulted in lower concentrations of propionic and butyric acids in rumen liquid than did the alfalfa-barley ration. The 90 per cent lowland hay ration resulted in less total VFA in rumen samples than did the control ration, and less butyric acid than rations containing lower levels of lowland hay. Concentrations of acetic acid were not affected by the different rations. Pelleting of the ration containing 45 per cent of lowland hay increased levels of total VFA because of increased concentrations of acetic, propionic and butyric acids. Pelleting of the other lowland hay rations had no significant effects on concentrations of

VFA. Rations containing 45 and 60 per cent of lowland hay resulted in higher proportions of acetic acid and lower proportions of propionic acid than did the alfalfa-barley ration. As the level of lowland hay in the ration was increased to 90 per cent, the proportion of acetic acid continued to increase, and the proportion of butyric acid decreased. Pelleting of the rations had no appreciable effects on the proportions of VFA in rumen liquid.

The second trial was undertaken to study the effects of pelleting of rations containing 60 and 90 per cent of lowland hay on concentrations and proportions of VFA in rumen samples obtained from fistulated wethers three hours after feeding. Samples were obtained, via the fistula, on the 1st, 7th, 14th, 21st and 28th days of the trial. It was observed that the alfalfa-barley rations resulted in higher concentrations of VFA in rumen fluid than did the lowland hay rations. Pelleting of the rations increased concentrations of total VFA, acetic, propionic and butyric acids in rumen liquid. Lower proportions of acetic acid, and higher proportions of butyric acid were found with the alfalfa-barley ration, than with the lowland hay rations. The percentage of propionic acid was not affected by the different rations. Pelleting of the rations had no effect on the proportions of VFA in rumen fluid.

The results of the experiment indicated that concentrations of VFA in rumen samples did not reflect differences obtained in feed intake as a result of pelleting the rations. Pelleting of the rations had no effect on proportions of VFA, and would not influence utilization of energy supplied as VFA. Consequently, analyses of VFA in rumen liquid were not an accurate measure of the value of a ration for productive purposes.

IV. Effects of Pelleting and Level of Lowland Hay on Cellulose Digestion in the Rumen

The trial was conducted to study the effects of pelleting of rations containing 60 and 90 per cent of lowland hay on cellulose digestion in the rumen. Cellulose digestion was measured by an in vivo technique whereby shredded filter paper in nylon bags was placed in the rumina of fistulated wethers for 12 and 24 hour periods. It was noted that non-pelleted rations resulted in greater cellulose digestion than did the alfalfa-barley ration. Increasing the level of lowland hay from 60 to 90 per cent of the ration reduced the percentage of cellulose digested. Pelleting of the 60 per cent lowland hay ration reduced cellulose digestion, whereas pelleting of the 90 per cent lowland hay ration increased cellulose digestion, as compared to similar non-pelleted rations.

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Appendix A. Statistical Treatment of Feeding Trial Data

Table I. Analyses of Variance

		Source of Variation	DF	M.S.	F
Trial 1	Daily Gain	Treatments	6	0.08	4.0**
		Error	49	0.02	
	Adjusted Daily Gain	Treatments	6	0.01	0.5
		Error	49	0.02	
Trial 2	Dry Matter/Day	Treatments	6	4.84	60.5**
		Error	21	0.08	
Trial 3	Daily Gain	Treatments	6	0.10	10.0**
		Error	35	0.01	
	Adjusted Daily Gain	Treatments	6	0.03	3.0*
		Error	35	0.01	
Trial 2	Digestion Coefficients				
	Dry Matter	Treatments	6	362.33	19.8**
		Error	21	18.28	
	Crude Protein	Treatments	6	148.00	22.0**
		Error	21	6.71	
	Gross Energy	Treatments	6	215.67	20.8**
		Error	21	10.38	

* Significant at the 5% level
** Significant at the 1% level

Table II. Significant Mean Differences*

Trial 1 — Daily Gain

Treatments	6	1	4	2	5	7	3
Means	0.34	0.35	0.35	0.38	0.51	0.51	0.53

Trial 2 — Dry Matter/Day

Treatments	6	4	2	7	1	5	3
Means	0.59	1.39	1.79	2.43	3.03	3.30	3.39

Trial 3 — Daily Gain

Treatments	2	4	6	1	7	5	3
Means	0.16	0.17	0.17	0.28	0.34	0.42	0.46

Adjusted Daily Gain

Treatments	4	2	6	7	3	5	1
Means	0.20	0.22	0.23	0.28	0.30	0.35	0.40

Trial 2 — Digestion Coefficients

Dry Matter

Treatments	6	7	3	5	4	2	1
Means	50	58	65	70	71	72	78

Crude Protein

Treatments	6	7	2	3	4	1	5
Means	57	63	70	70	71	73	75

Gross Energy

Treatments	6	7	3	5	4	2	1
Means	57	58	64	69	70	71	76

* Means not underscored by the same line are significantly different at the 5% level

Appendix B. Manometric Fermentation Procedure

Approximately 50 ml. of each rumen sample was homogenized five minutes in a Virtis "23" homogenizer, under a stream of carbon dioxide to maintain anaerobiosis. The homogenized sample was strained through two layers of cheesecloth and 1 ml. of liquid was pipetted into each Warburg flask (17 to 19 ml. capacity). Carbon dioxide was passed rapidly through a sodium bicarbonate buffer solution (Hungate, 1950) for one minute to adjust pH to slightly below 7.0, and 2 ml. of the buffer was added to each flask. 0.3 ml. of 6N H_2SO_4 was added to the side arm of each flask, and the flasks were attached to manometers and placed in the water bath at 40^oC.

Carbon dioxide gas, at a pressure of at least 20 mm. Hg. above atmospheric, was passed through the flasks, three minutes with shaking and four minutes without shaking, to ensure displacement of all air and attainment of equilibrium between liquid and gas phases. The carbon dioxide flow was then shut off, and gas sucked out of the manometer and flask until the mercury level in the distal arm of the manometer measured 25 mm. when the level in the proximal arm measured 180 mm. Flasks were shaken 10 minutes to equilibrate to the new pressure. Pressure readings were taken at the end of this period, and every 10 minutes thereafter for one hour. Approximately one hour elapsed between sample collection and the time when the first measurements were taken.

Two control and two experimental flasks were incubated for each rumen sample to provide duplicate determinations. Acid in the side-arm of each control flask was added to the fermentation medium at the beginning of the incubation to liberate 'initial' carbon dioxide in the bicarbonate, and the resulting pressure recorded. After one hour of incubation the 'fermentation' pressure in each experimental flask was recorded, acid was tipped in from the side-arm, and 'total' pressure

determined. The mercury level in the proximal arm of the manometer was maintained at 180 mm. throughout the incubation period, and pressure measurements were taken on the distal arm.

Each pressure noted above was multiplied by the appropriate flask constant (Umbreit *et al.*, 1957) to determine µl. gas produced. Acid production was calculated as follows:

(1) 'Total' gas - 'fermentation' gas = 'residual' gas
(Gas left in the bicarbonate after one hour of incubation).

(2) 'Initial' gas - 'residual' gas = gas liberated by acid produced in the fermentation.

Appendix C. Statistical Treatment of Manometric Fermentation Data

Table I. Analyses of Variance

Source of Variation	DF	Total Gas Produced M.S.	F	Total Acid Produced M.S.	F
Trial 1					
Treatments	6	9726.01	21.53**	4.41	14.70**
Sampling Time	3	2553.63	5.65**	1.59	5.30**
Interaction	18	386.11	0.85	0.24	0.80
Error	80	451.64		0.30	
Trial 2					
Treatments	4	1538.26	8.96*	1.12	3.11
Replicates	1	210.68	1.23	0.61	1.69
Error	4	171.63		0.36	

* Significant at the 5% level
** Significant at the 1% level

Table II. Significant Mean Differences*

Trial 1

Total Gas Production

Treatments	6	7	4	5	3	2	1
Means	11.5	15.2	54.2	56.3	59.0	59.6	83.1
Sampling Time	0	6	1½	3			
Means	35.7	51.1	54.7	57.6			

Total Acid Production

Treatments	6	7	4	2	3	5	1
Means	0.18	0.28	1.08	1.17	1.18	1.20	1.73
Sampling Time	0	6	1½	3			
Means	0.68	0.96	1.13	1.24			

Trial 2

Total Gas Production

Treatments	7	6	4	5	1
Means	10.8	12.6	32.3	33.2	79.6

* Means not underscored by the same line are significantly different at the 5% level

Table III. T-Test' for Differences Between Comparable Treatment Means in Trials 1 and 2

Treatments	1	4	5	6	7
Pressure Increase					
t	1.05	3.30	1.56	0.91	3.10
$P_{.05}$*	3.81	6.86	11.75	11.27	7.47
Acid Production					
t	0.33	0.56	0.80	1.60	0.00
$P_{.05}$	2.78	2.78	2.78	3.18	2.78

' Goulden (1956).

* Value of t required for significance at the 5% level when $n_1 \neq n_2$ and variances are assumed different

Appendix D. Statistical Treatment of Data on Volatile Fatty Acids in Rumen Fluid

Table I. Analyses of Variance: Trial 1

Source of Variation	D.F.	Total VFA		Acetic Acid		Propionic Acid		Butyric Acid		Valeric and Higher Acids	
		M.S.	F	M.S.	F	M.S.	F	M.S.	F	M.S.	F
A. Molar Concentrations											
Treatments	6	13.83	5.18**	2.98	2.52*	1.52	10.13**	1.45	12.08**	0.077	4.05**
Time of Sampling	3	36.52	13.68**	14.45	12.24**	1.85	12.33**	0.85	7.08**	0.010	0.53
Interaction	18	3.88	1.45	1.59	1.35	0.22	1.47	0.14	1.17	0.021	1.10
Error	80	2.67		1.18		0.15		0.12		0.019	
B. Molar Proportions											
Treatments	6			347.03	23.21**	108.45	11.09**	112.90	12.30**	6.24	1.43
Time of Sampling	3			15.97	1.07	14.65	1.50	4.60	0.50	13.90	3.20**
Interaction	18			22.94	1.53	16.22	1.66	5.19	0.56	2.94	0.68
Error	80			14.95		9.78		9.18		4.35	

*Significant at the 5% level
**Significant at the 1% level

Table II. Significant Mean Differences in Molar Concentrations: Trial 1*

Total VFA	Treatments	7	6	2	4	5	1	3
	Means	5.91	6.26	6.47	6.73	7.65	7.89	8.44
	Sampling Time	0	6	1½	3			
	Means	5.54	6.82	7.93	8.02			
Acetic Acid	Treatments	2	7	4	1	6	5	3
	Means	4.16	4.19	4.43	4.46	4.54	5.03	5.32
	Sampling Time	0	6	1½	3			
	Means	3.60	4.49	5.06	5.22			
Propionic Acid	Treatments	7	6	4	5	2	3	1
	Means	1.07	1.14	1.21	1.33	1.33	1.55	1.99
	Sampling Time	0	6	3	1½			
	Means	1.06	1.28	1.59	1.60			
Butyric Acid	Treatments	6	7	2	4	5	1	3
	Means	0.46	0.51	0.79	0.80	1.06	1.14	1.28
	Sampling Time	0	6	3	1½			
	Means	0.64	0.85	1.00	1.02			
Valeric and Higher Acids	Treatments	6	7	2	5	3	4	1
	Means	0.12	0.14	0.19	0.23	0.28	0.29	0.30

* Means not underscored by the same line are significantly different at the 5% level

Table III. Significant Mean Differences in Molar Proportions: Trial 1*

Acetic Acid	Treatments	1	3	2	5	4	7	6
	Means	57.7	62.6	64.4	65.8	66.1	70.5	72.3
Propionic Acid	Treatments	5	4	7	6	3	2	1
	Means	17.3	17.9	17.9	18.3	18.8	20.1	24.9
Butyric Acid	Treatments	6	7	4	2	1	5	3
	Means	7.3	8.7	11.9	12.2	13.7	13.8	15.0
Valeric and Higher Acids	Sampling Time	3	6	1½	0			
	Means	2.7	3.0	3.1	4.3			

* Means not underscored by the same line are significantly different at the 5% level

Table IV. Analyses of Variance of Molar Concentrations: Trial 2

Source of Variation	D.F.	Total VFA		Acetic Acid		Propionic Acid		Butyric Acid		Valeric and Higher Acids	
		M.S.	F	M.S.	F	M.S.	F	M.S.	F	M.S.	F
A. Pretreatment Period											
Days on Test	1	7.73	1.92	2.57	2.86	0.02	0.08	0.41	0.74	0.16	3.20
Wethers	4	5.90	1.47	1.22	1.36	0.75	3.00	0.24	0.44	0.05	1.00
Error	4	4.02		0.90		0.25		0.55		0.05	
B. Trial Period											
Treatments	4	51.00	13.82**	12.08	8.95**	2.72	9.38**	3.13	16.47**	0.142	10.14**
Days on Test	4	22.98	6.23**	9.67	7.16**	1.12	3.86*	0.44	2.32	0.032	2.28
Replicates	1	35.66	9.66**	16.96	12.56**	1.17	4.03	0.58	3.05	0.000	0.00
Treat. x Days	16	6.34	1.72	2.37	1.76	0.31	1.07	0.29	1.53	0.021	1.50
Treat. x Rep.	4	18.99	5.15**	7.96	5.90**	1.00	3.45*	0.48	2.53	0.052	3.71*
Days x Rep.	4	2.81	0.76	1.02	0.76	0.40	1.38	0.31	1.63	0.072	5.14**
Triple Interaction	16	3.69		1.35		0.29		0.19		0.014	

* Significant at the 5% level
** Significant at the 1% level

Table V. Significant Mean Differences in Molar Concentrations: Trial 2*

Total VFA

Treatments	6	4	7	5	1					
Means	7.76	9.18	9.19	11.86	13.31					
Days on Test	1	7	14	28	21					
Means	8.20	9.16	11.06	11.08	11.82					
Replicates	1	2								
Means	9.42	11.11								
Treat. x Rep.	7-1	6-1	6-2	4-2	5-1	4-1	7-2	1-1	1-2	5-2'
Means	7.45	7.65	7.88	8.42	9.15	9.94	10.94	12.90	13.72	14.57

Acetic Acid

Treatments	6	4	7	5	1					
Means	5.00	5.71	5.83	7.31	7.54					
Days on Test	1	7	14	28	21					
Means	4.87	5.66	6.76	6.83	7.26					
Replicates	1	2								
Means	5.69	6.86								
Treat. x Rep.	7-1	6-1	6-2	4-2	5-1	4-1	7-2	1-1	1-2	5-2'
Means	4.78	4.88	5.11	5.28	5.44	6.14	6.87	7.22	7.85	9.18

Propionic Acid

Treatments	6	4	7	5	1					
Means	1.63	1.94	2.07	2.46	2.99					
Days on Test	1	7	14	28	21					
Means	1.79	1.98	2.26	2.44	2.61					
Treat. x Rep.	7-1	6-1	6-2	4-2	4-1	5-1	7-2	1-2	5-2	1-1'
Means	1.52	1.54	1.73	1.84	2.04	2.06	2.63	2.81	2.85	3.16

Butyric Acid

Treatments	6	7	4	5	1
Means	0.87	1.04	1.27	1.71	2.25

Valeric and Higher Acids

Treatments	7	6	4	5	1					
Means	0.25	0.27	0.27	0.38	0.53					
Treat. x Rep.	6-2	7-1	4-2	5-1	7-2	4-1	6-1	5-2	1-2	1-1'
Means	0.19	0.21	0.23	0.28	0.30	0.31	0.35	0.48	0.52	0.54
Days x Rep.	28-1	1-2	7-2	7-1	14-2	14-1	21-1	28-2	21-2	1-1#
Means	0.22	0.25	0.25	0.27	0.32	0.36	0.37	0.45	0.46	0.48

*Means not underscored by the same line are significantly different at the 5% level.

'Treatment 5 in Replicate 2 and Treatment 1 in Replicate 1.

#1st Day on test in Replicate 1.

Table VI. Analyses of Variance of Molar Proportions: Trial 2

Source of Variation	D.F.	Acetic Acid M.S.	Acetic Acid F	Propionic Acid M.S.	Propionic Acid F	Butyric Acid M.S.	Butyric Acid F	Valeric and Higher Acids M.S.	Valeric and Higher Acids F
A. Pretreatment Period									
Days on Test	1	0.10	0.009	10.00	2.49	3.02	0.24	3.02	0.88
Wethers	4	10.67	0.94	10.30	2.56	4.54	0.36	0.66	0.19
Error	4	11.30		4.02		12.57		3.42	
B. Trial Period									
Treatments	4	86.80	9.99**	3.56	0.33	45.72	4.98**	1.28	0.55
Days on Test	4	11.91	1.37	5.38	0.50	6.96	0.76	10.90	4.66*
Replicates	1	20.48	2.36	0.19	0.02	2.21	0.24	12.10	5.17*
Treat. x Days	16	5.81	0.67	13.68	1.28	6.91	0.75	1.74	0.74
Treat. x Rep.	4	4.14	0.48	39.64	3.71*	20.66	2.25	1.13	0.48
Days x Rep.	4	13.10	1.51	29.96	2.80	20.43	2.23	14.00	5.98**
Triple Interaction	16	8.69		10.69		9.17		2.34	

*Significant at the 5% level
**Significant at the 1% level

Table VII. Significant Mean Differences in Molar Proportions: Trial 2*

Acetic Acid

Treatments	1	5	4	7	6
Means	56.7	61.3	62.0	62.9	64.5

Propionic Acid

Treat. x Rep.	7-1	5-2	6-1	1-2	4-1	6-2	4-2	5-1	7-2	1-1'
Means	18.9	19.6	19.9	20.4	20.5	21.6	22.4	23.5	24.0	24.5

Butyric Acid

Treatments	6	7	4	5	1
Means	11.2	12.3	13.6	13.6	16.9

Valeric and Higher Acids

Days on Test	7	28	14	21	1					
Means	2.8	2.9	3.0	3.3	5.3					
Replicates	2	1								
Means	3.0	4.0								
Days x Rep.	7-2	28-1	14-2	1-2	21-1	14-1	7-1	28-2	21-2	1-1#
Means	2.3	2.3	2.8	2.8	3.1	3.3	3.3	3.5	3.5	7.8

*Means not underscored by the same line are significantly different at the 5% level.
'Treatment 1 in Replicate 1.
#1st Day on test in Replicate 1.